IV. *Experimental Researches on Vegetable Assimilation and Respiration. III.—On the Effect of Temperature on Carbon-Dioxide Assimilation.**

By Gabrielle L. C. Matthaei, *Fellow of Newnham College, Cambridge.*

Communicated by F. Darwin, *F.R.S.*

Table of Contents.

* The first two papers of this series emanating from the Cambridge Botanical Laboratory were written by Dr. F. F. Blackman and will be found in vol. 186 (1895) of the 'Philosophical Transactions,' Series B, of the Royal Society.

Section I.—Introduction and Historical Survey.

Introduction.

THAT the activity of a plant in the function of assimilation is influenced to a certain extent by the temperature to which it is subjected, has long been recognised, but, in spite of the universal acceptance of this principle, comparatively little detailed study has been undertaken in confirmation of it.

The present investigation was undertaken at the suggestion of Dr. F. F. BLACKMAN and has been carried out in his laboratory. The problem proved to be much more complex than was anticipated and I have to acknowledge, with great pleasure, my indebtedness to Dr. BLACKMAN for frequent valuable advice and several important suggestions.

As the nature of the relation under investigation only unfolded itself gradually in the actual course of the work, I have thought it permissible to expound it in the same way. It will be well, therefore, that the reader should bear in mind that the earlier experiments are, in some cases, of rather a provisional nature though, I trust, well worth permanent record.

This research differs from any which has preceded it on the subject, in the precautions adopted for determining the true internal temperature of the leaf, and in the attention paid to constancy in the environment of the leaves *before* the experiment. A new leaf was used for every experiment, a large number of leaves being kept ready under uniform conditions.

It has always been recognised that to study the effect of a single definite condition on the assimilation or respiration, the other conditions affecting the process must be kept as uniform as possible. Now an additional source of complications has been recognised, for recent investigations by Dr. BLACKMAN and myself have shown that these functions are influenced, not only by the external conditions at the actual time of experiment, but also very considerably, by internal conditions which are the result of the external conditions to which the plant has been subjected for some time previously. Such factors, the effect of which persists, are starvation, excess of nutrition and temperature changes. Thus it is not sufficient to place plants under constant conditions, their whole previous treatment must also have been similar. The non-recognition of this after-effect necessarily vitiates the results of many of the older investigations, in which a return to the normal state was assumed, provided the plant appeared uninjured.

I shall consider here at some length the work of KREUSLER (1) (1888), both because it affords a good illustration of the errors introduced by neglecting uniformity in previous conditions, and also because this investigation has hitherto been accepted as giving the true relation between temperature and assimilation.

(i) *Consideration of the Researches of Kreusler.*

The greater part of this long and detailed investigation is devoted to the determination of the assimilation and respiration, at various temperatures, of a shoot of *Rubus*. The same shoot was used throughout, and was never removed from the glass-fronted metal box in which the experiment was conducted.

To the back of this box was attached a cell of circulating water by means of which the temperature of the chamber was automatically regulated. This arrangement seems to have been satisfactory only at medium temperatures. Both at the higher and lower limits, a great difference of temperature, sometimes amounting to 5° C., occurred between the back of the chamber and the front. KREUSLER took the mean temperature as the true one, but, to take an actual case, the effect of keeping some leaves at 0° C. and some at 5° C. can hardly be represented by supposing them all to be at 2°·5 C. At the higher temperatures, where increase of a degree or two might be fatal, this approximation is even less satisfactory.

The light used was that of the electric arc. An air current containing a known percentage of CO_2 was drawn through the plant-chamber, and the amount of CO_2 consumed by the plant was estimated gravimetrically, by absorption of the residual CO_2 by baryta.

Assimilation readings of one hour and respiration readings of several hours were taken at the various temperatures. The assimilation (apparent) and respiration at any one temperature were not estimated consecutively, but whenever convenient— sometimes after the lapse of some days, during which other experiments had been performed on the plant.

KREUSLER summarised his results in the form of a curve,* taking the mean of all assimilation readings at a given temperature, though these were sometimes extremely divergent. He found that the assimilation rose rapidly from 2°·3 C.–11°·3 C. After this point the assimilation increased very slowly, attaining a maximum value at 25° C. Then the assimilatory curve fell again, until the faculty of assimilation was destroyed at the higher temperatures.

The investigations by KREUSLER are an enormous advance on any previous work. His estimations are most careful and no doubt accurate, but his treatment of the plants themselves calls for criticism and especially his non-recognition of the effect of previous conditions.

A leaf, which has been exposed to a temperature above 30° C., may not show any outward marks of injury, but its respiration and assimilation at some medium temperature, such as 18° C., will be very different from what they would have been had it never been exposed to the higher temperature.

As evidence of this point, the following details of an experiment performed at the outset of my own investigations may be given :—

* See PFEFFER (2), ' The Physiology of Plants,' vol. 1, Section 58.

The respiration of twelve cherry-laurel leaves was estimated during 12 hours at 17° C., and then the assimilation of one of them was taken at the same temperature, giving a value for the real assimilation of ·0037½ gramme CO_2 per hour. After some hours' exposure to 30° C., determinations with the same leaves at 21° C. gave a value for the assimilation of only ·0018½ gramme CO_2 per hour. This could not be the true effect, for the amount is about half that obtained at the neighbouring temperature of 17° C. Evidently the leaves had suffered by their exposure to the high temperature, and the assimilation could not afterwards rise above a certain limit.

The importance of this experiment lies in its close resemblance to the procedure adopted by KREUSLER, throwing a very unfavourable light on the trustworthiness of his results.

Further we must consider the effect of nutrition on these functions of respiration and assimilation.

It has been shown both by BOUSSINGAULT (3) and by SAPOSCHNIKOFF (4), that, if a leaf be allowed to assimilate actively for some time, its power of assimilation may diminish, owing to the accumulation of reserve material, which in some way dislocates the machinery. Such an effect, however, is only produced after prolonged assimilation and would not affect KREUSLER's results, since his assimilation experiments were only an hour in duration.

Much more important is the effect of the nutrition on the respiration. Prolonged darkness produces a steady diminution in the amount of CO_2 given off by a leaf; but if such a starved leaf be allowed to assimilate actively even for an hour or so, the respiration will at once increase greatly, although much additional reserve material may not have been produced. Thus the respiration of a leaf does not depend merely on the temperature, but is greatly influenced by recent assimilation or starvation BORODIN (5).

If we consider the investigations of KREUSLER, we see that he entirely ignored these factors. He assumed that the branch of *Rubus* was throughout in a normal condition and was completely undisturbed by the various changes of temperature or of nutrition which it had undergone. For this reason, he is quite content to estimate the assimilation (apparent) at a certain temperature on one day, and the respiration at that temperature several days later, meanwhile starving and feeding the leaves at will by other respiration and assimilation experiments and exposing them to very various temperatures. Every estimation must have been affected by those which preceded it, and this effect would have been cumulative, making each estimation more untrustworthy than the one before.

A consideration of KREUSLER's numbers will give the best proof of the truth of this criticism.

It will be seen, on looking through the experiments, that only once was a second estimation taken of the respiration at any temperature, *i.e.*, at 33° C. on two

consecutive days. The assimilation at 33° C. was also taken twice, on the same days as the respiration. The following are the data obtained :—

August 8, morning assimilation, 53·4 ; afternoon respiration, 35·4.

August 9, morning assimilation, 43·8 ; afternoon respiration, 30·4.

These numbers are most striking, for although these experiments were performed on consecutive days and the plant was subjected to no injurious temperature in the interval, yet there is a most unmistakable falling-off on the second day, both in the assimilation and in the respiration, showing that the exposure to the temperature of 33° C. on the first day had most certainly affected the vitality of the plant. This effect would most probably persist during further experiments on the branch.

If all amounts of assimilation given in his paper be plotted out (fig. 1) in their

Fig. 1.
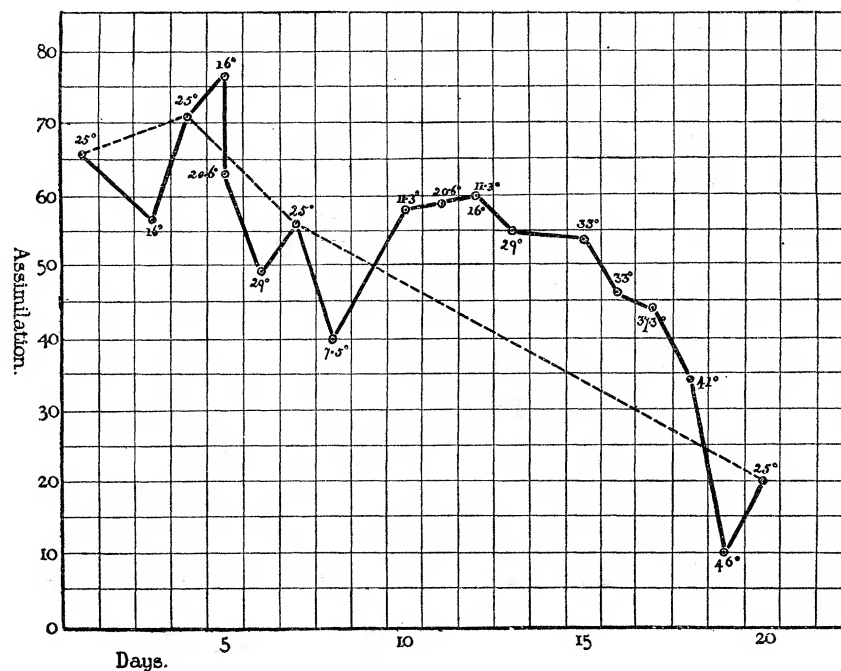
chronological order, without regard to the temperatures, a curve is obtained, which is well worth detailed consideration. The general character is most decidedly that of a progressive fall in the amount of the assimilation, notwithstanding that the various temperatures occur in no particular order, but quite capriciously, and that the same temperature is often repeated. Thus, in this series, the amount of assimilation performed by the *Rubus* branch was largely a function of the time during which the experiment had lasted, and was not purely dependent on the temperature.

Moreover, it is most striking that all the highest readings occur during the first few days, and before the plant was subjected to any temperature higher than 25° C. ;

thus the assimilation numbers for the first days are 65, 67, 71, 77, 63 ; they never rise as high as 60 after the plant has been at the temperature of 29° C., although the self-same temperatures are again tried. At the beginning also occur the greatest oscillations in the curve, showing that the plant is there able to respond adequately, as regards assimilation, to variations in temperature.

After the eighth day there is a change ; the amounts of assimilation at 29° C., 20° C., 11° C., 16° C., and 33° C., all have about the same value—indeed, they agree among themselves far better than do readings of the same temperature at different times, from which we again see the influence of the time-effect. Now come the experiments at 33° C., which have been already mentioned, and which are the prelude to a very rapid fall in the assimilation. The fact that subsequent experiments were all at temperatures above 33° C. easily explains this, for the " after-effect " is greater the higher the temperature. The leaf was kept for 8 hours at 37° C., and for 8 hours at 42° C., but nevertheless was still considered to be in a normal state for assimilation at 46° C.

As a finale to the series, the assimilation was again taken at 25° C. and found to be 23·9, in contrast to the values of 65·7, 71 and 56·1 obtained before. This enormous fall must indicate a very great diminution in the vitality of the leaf. KREUSLER, however, was quite content, because it was greater than the assimilation at 46° C., and showed that the branch could still respond to the stimulus of change of temperature. The four values obtained for the assimilation at 25° C., if plotted out in their chronological order (as dotted in fig. 1), probably give a very fair indication of the fall in vitality of the leaf.

It will be obvious that KREUSLER's results are not in the least the expression, as he conceived them to be, merely of the effect of the temperature on the assimilation, but are the outcome of the most various causes, so complicated, that it is impossible to extricate from them the simple temperature effect.

(ii) Consideration of the Researches of Other Investigators.

The remaining investigations, except those of HEINRICH, amount to little more than a general confirmation of the fact that temperature does affect assimilation and more or less isolated determinations of the lowest temperature at which this function persists.

CLOËZ and GRATIOLET (6) (1851) found that for water-plants, the lowest temperature at which assimilation would take place, was 10–15° C.

As regards land plants BOUSSINGAULT (7) (1867) found that oxygen was produced by *Pinus laricio* and by certain grasses at temperatures of −0°·5 C. to +3°·5 C., from which he concludes that green trees and the grass of the meadows may very well assimilate throughout the winter.

No importance need be attached to the work of FRANCOPERT (8) (1864), who

attempted to express the connection between the assimilation and the temperature by a mathematical formula. In this research the distinction between the respiration and assimilation was ignored.

BOEHM (9) (1873), in the course of a long paper on intramolecular respiration of leaves, gives the results of a few determinations of the assimilation of *Juglans* at different temperatures, but these are of no great interest.

More important is the work of HEINRICH (10) (1871) on *Hottonia palustris*. His paper embodies the result of a careful comparative study of the effect of different temperatures on the oxygen production of the plant. Unfortunately his results are vitiated by the method he employed, that of counting the number of bubbles evolved in the light from the cut end of the stalk.

Avoidable sources of error, such as the expansion of the gas due to the rise of temperature, movements in the water, &c., are very clearly considered in HEINRICH's paper, together with the precautions taken to overcome them. There are, however, certain defects inherent in this method* which cannot be remedied by any amount of care. The most important of these are the following :—(1) solution of the oxygen in the water, on account of which the absence of gas formation at a low temperature is no proof that assimilation has entirely ceased ; (2) the apparent assimilation (*i.e.*, real assimilation less the respiration) only is obtained, this error having most effect the higher the temperature ; (3) the same plant must be used throughout since the rate of bubbling depends on the aperture, &c., and thus the experiments are not all performed under exactly similar conditions, since the intermediate treatment of the plant has varied.

HEINRICH found that the assimilation of *Hottonia palustris* increased rapidly with the temperature, reaching its optimum at 31° C., and then fell on a further increase of temperature, until it ceased between 50° C. and 62° C. If the numbers given for the rate of bubbling be erected as the ordinates on the temperature as abscissa, a curve is obtained, which is exactly symmetrical in its rise and fall, and of which the highest point, *i.e.*, maximal assimilation, is the mean point of the temperature range.

This curve is founded on the apparent assimilation, and for this reason only the first part of the curve can represent the effect of the temperature. The higher the temperature the more the results would be vitiated by the neglect of the respiration and by the progressive enfeeblement of the plant.

At the end of the paper, HEINRICH considers the question of the limiting temperatures for assimilation. Intermittent bubbles were formed at 3°·1 C., but a continual stream not below 5°·6 C. This limiting temperature is very much lower than that found by CLOËZ and GRATIOLET (6), although water plants were used in both cases.

PRJANISCHNIKOW (13) (1876) is the only observer who has taken into consideration

* There is a full discussion of the disadvantages of this method in papers by PFEFFER (11) and REINKE (12).

the effect of varying the intensity of the light at different temperatures. For *Typha latifolia* he found that in direct sunlight the temperatures between 9°·5 C. and 39° C. all gave the same result, but that in diffuse light the assimilation increased in passing from 7–17° C., but fell off in passing from 17–25° C. He concludes that in strong light, when the assimilation is already very great, the temperature cannot increase it further. The falling off of the CO_2 assimilation in diffuse light with temperatures between 17–25° C. he explained by the increase in respiration. These results are quite contrary to those obtained in the present research.

VAN TIEGHEM (14) 1892 in the six lines devoted to this subject in his text-book* expresses the opinion that assimilation bears the same sort of relation to temperature as does respiration. He supports this by quoting four, then (as now) unpublished numbers by BONNIER and MANGIN, which show that the oxygen production of a leafy elm branch augments with increasing rapidity as the temperature rises, at least up to 30° C. Slight and incomplete as it is, this statement gets nearer to the truth than any other published.

There is but one further paper to which reference must be made. In the detailed account given by JUMELLE (15) (1892) of the assimilation and respiration of the lichens, the results of a few experiments performed on individuals of this class at exceedingly low temperatures are given, and, for the purposes of comparison, also a few experiments on phanerogams.

JUMELLE, who was the first to experiment below 0° C., claims to have proved that respiration ceases about −10° C., whereas assimilation persists to −30° C. or even to −40° C. His procedure is, however, unsatisfactory.

A limited amount of CO_2 was admitted into the plant chamber through a long glass inlet tube ; samples of the air were then removed through the same tube at the beginning and end of the experiments and were analysed by means of a Bonnier and Mangin apparatus.

In the first place, exception must be taken to the long inlet tube, and to the fact that it serves both to admit the CO_2 and to remove the samples. It might be some time before identity of composition between the air in the chamber and the air in the tube was established by diffusion, and the air removed for analysis would consist mainly of the contents of the tube. Since to indicate respiration the second sample must contain more CO_2 than the first, the existence of a feeble respiration might easily be masked by such an experimental error. On the other hand, a feeble assimilation might be augmented by the same cause.

JUMELLE's results suggest exactly such errors, respiration ceasing at a temperature far above that at which assimilation continued. Blank experiments performed under the same conditions would show at once whether this criticism is just or not. In the absence of any indication as to the probable experimental errors, and considering the impossibility of comparing the relative amounts of respiration and assimilation, we must at present reserve our acceptance of these results.

* 'Traité de Botanique,' vol. 1, p. 179.

In the second place the assimilation result appears to be based on the solitary case of the lichen *Evernia prunastri*. The two lichens of which the respiration was shown to have ceased at $-10°$ C., did *not* assimilate at $-30°$ C. and $-40°$ C. That they would assimilate at $-10°$ C., JUMELLE assumed but did not prove. The respiration of the two phanerogams, *Picea* and *Juniperus*, which assimilated at $-30°$ C. and $-40°$ C., was not taken at all (at least there is no mention of it in the paper), so that these two cases cannot be taken as evidence of the persistence of assimilation beyond respiration. There remains, therefore, *Evernia prunastri* as the solitary instance in which respiration ceased at $-10°$ C., and assimilation could be detected at $-40°$ C.

Section II.—METHODS AND PROCEDURE.

(i) *Methods of Estimation and Apparatus.*

The apparatus employed in these investigations was designed by Dr. F. F. BLACKMAN, and I must here express my thanks to him for putting it at my disposal.

In this apparatus the amount of carbon dioxide withdrawn by a single leaf from a current of air containing carbon dioxide is taken as the measure of assimilation. The carbon dioxide is estimated volumetrically by absorption in a known volume of baryta, which is subsequently titrated against standard hydrochloric acid.

As a description of the apparatus has, as yet, not been published, I shall enumerate shortly the points necessary to the understanding of the data given later.

Two aspirators are, by an automatic device, kept dropping at exactly equal rates. The single current of air, enriched with the required amount of CO_2,[*] which enters the apparatus is, by their action, divided into two perfectly equal half-currents. One of these currents is led directly through a Pettenkofer tube ; the other first passes through the leaf chamber. The difference between the CO_2 shown by the two tubes at the end of the experiment gives the amount assimilated by the leaf. By this exact automatic halving of the current and double estimation, moderate variations in the *absolute* amount of CO_2 supplied cease to be an undeterminable source of error, as the control tube continually gives an exact measure of them.

The whole apparatus contains six pairs of Pettenkofer tubes, and the currents are shifted by clockwork from one pair to the next at intervals of 1, 2, 3 or 4 hours as desired. The apparatus can be left to take care of itself for six consecutive periods, and so assimilation experiments may be carried on continuously day and night. A recording apparatus is attached to the aspirators which gives graphic evidence whether any failure of equalisation has taken place during the time they have not been under observation.

The Pettenkofer tubes that have served their turn may be removed at any

* By the method described in the first paper of this series, 'Phil. Trans.,' B, vol. 186 (1895), p. 496.

convenient time and estimated by washing out their contents into a beaker and titrating with standard HCl and phenolphthalein. They are then refilled with a known amount of standard baryta solution and replaced in the apparatus until the clockwork brings the current round to them again.

Instead of attempting to exclude all traces of atmospheric CO_2 in this procedure, we have tried and found much easier and quite accurate the alternative course of making a definite allowance, "the washing factor," for the CO_2 contamination during washing, titrating and refilling. This has been arrived at by a large number of controls made for the purpose, and is a reasonably constant amount.

The aspirators were always adjusted to the rate of 800 cub. centims. per hour. The percentage of CO_2 contained in the air current varied between ·8 and 2·8, the exact amount in any experiment can be calculated from the amount of CO_2 given by the control tube per hour (see p. 87). Care was taken in each experiment that the amount supplied was greatly in excess of the highest amount that would be assimilated in that experiment, in order that there might not be the slightest lowering of the assimilation due to an insufficient supply of CO_2.

In the data given of all the experiments there will be two columns, one giving the residual amount of CO_2 in the current from the plant chamber, and the other, "the control," giving an amount of CO_2 equal to that supplied to the leaf chamber. The difference between these amounts will be the "apparent" assimilation.*

The successive numbers in any column of control estimations will show a general agreement with slight individual differences, but, as pointed out already, these affect both plant current and control current equally.

I give first the results of two blank experiments, the difference between the amounts of carbon dioxide found in the corresponding tubes is a measure of the total experimental error, including possible slight leaks in the apparatus, titration errors, &c. The procedure exactly resembled that of an ordinary experiment, but

Experiment I.—January 14–15.

Time.	All the amounts are expressed in grammes of CO_2.		
	Control tubes.	Tubes from chamber.	Difference.
8.30–10.30 P.M. . .	Preliminary	Preliminary	—
10.30–12.30 A.M. . .	·0307$\frac{1}{2}$	·0305$\frac{1}{2}$	+ ·0002
12.30– 2.30 ,, . .	·0305$\frac{1}{4}$	·0305	+ ·0000$\frac{1}{4}$
2.30– 4.30 ,, . .	·0305$\frac{1}{4}$	·0304	+ ·0001$\frac{1}{4}$
4.30– 6.30 ,, . .	·0303$\frac{1}{2}$	·0304$\frac{1}{2}$	– ·0001
6.30– 8.30 ,, . .	·0298$\frac{1}{2}$	·0299	– ·0000$\frac{1}{2}$

* The "apparent" assimilation is, of course, numerically less than the *real* assimilation by the amount of respiratory CO_2 that the leaf is to be considered as producing (in the light) at the moment under consideration.

EXPERIMENT II.—April 15–16.

Time.	All amounts are expressed in grammes of CO_2.		
	Control tubes.	Tubes from chamber.	Difference.
10.30–12 noon . .	Preliminary	Preliminary	—
12– 2 P.M.	·0321½	·0322½	− ·0001
2– 4 „	Spoilt	—	—
4– 6 „	·0317½	·0316½	+ ·0001
6– 8 „	·0315	·0310½	+ ·0004½
8–10 „	·0309	·0313	− ·0004
10–12 midnight . .	·0312	·0315	− ·0003
12– 2 A.M.	·0315	·0315½	− ·0000½
2– 4 „	·0317½	·0318½	− ·0001
4– 6 „	·0316	·0317	− ·0001
6– 8 „	·0313½	·0312½	+ ·0001
8–10 „	·0313	·0312½	+ ·0000½

there was no plant in the chamber. It will be seen that the numbers in the "difference" column agree much more closely than the absolute amounts of CO_2, proving the exact halving of the current.

The small value of the experimental errors shown in these experiments, and the good agreement of the assimilation numbers subsequently to be given, prove that a very fair degree of accuracy in the CO_2 determinations has been obtained.

This method of continuous observation possesses most important advantages. Among others every determination of the assimilation depends not on a single estimation, but on four or five consecutive estimations made under unchanged conditions, and this is of immense value in an investigation of this nature in which the experimental difficulties are great, and the factors which may affect the result numerous.

For the assimilation experiments, a specialised form of flat glass leaf-chamber was employed. This consisted of a broad rim of iron somewhat larger than the outline of the leaf, with an exit tube through the top and an inlet tube through one side near the base. A plate of glass fastened permanently to one surface of this rim by wax formed the front of the chamber. The back was formed by a similar piece of glass which was waxed on to the other surface of the rim each time after the leaf was put in, care being taken that the whole should be quite air-tight. This made a shallow chamber about 5 millims.
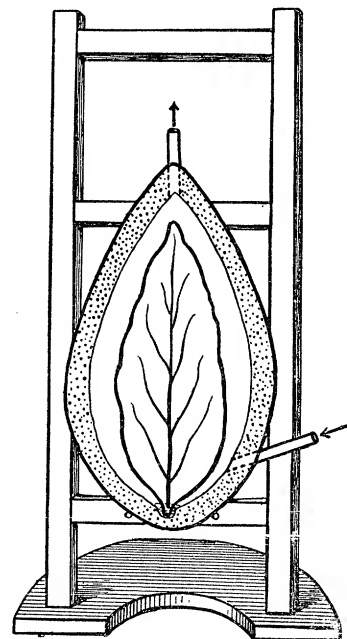
Leaf-chamber on stand.

deep, provided with inlet and outlet tubes. The content of such a chamber is not more than 40 cub. centims.

The chamber was fastened vertically to a brass stand. As the chamber was very shallow, the leaf remained flat and erect, lightly held between the back and the front glasses. The stalk dipped into a small amount of water.

The leaf chamber with its stand was placed in a large rectangular copper bath, filled with water, the front of the bath having a glass window, so that the leaf may be illuminated.

For respiration experiments involving several leaves, larger vessels of metal were employed.

(ii) *Regulation of the Temperature.*

For high temperatures the bath was heated by a small flame controlled by a thermostat. Temperatures near that of running water (5–14° C. according to the time of year) could be obtained by allowing water to circulate freely through the bath. Intermediate temperatures, *i.e.*, those between those of running water and the higher temperatures, could be obtained by combining both the flame and the current of water.

By these means the temperature of the bath could be maintained very constant, the variation during any individual experiment seldom amounting to more than 0·2–0·4° C. of a degree.

The air current before entering the leaf chamber always passed through a long spiral tube immersed in the bath, in order that it might attain the exact temperature of the bath.

There was some difficulty in keeping the temperature of the bath constant in the neighbourhood of 0° C. and below it, especially on account of the proximity of the incandescent burners used as source of illumination.

For temperatures just above 0° C., the bath was filled with a mixture of ice and water, for temperatures below 0° C., with a mixture of ice and salt solutions of various strengths. Ice was continually added and solid salt from time to time to remedy the dilution produced by the ice. Although the ice floats at the top, a uniform temperature throughout the bath was obtainable by the energetic use of an automatic air-stirrer.

The continual addition of ice and salt soon produces a slight opalescence in the bath-solution in spite of washing the ice and filtering the salt solution, but a consideration of Table IV. shows that the effect on the results has been unappreciable, as there is no falling-off during the course of an experiment, although the opalescence always increased with the time.

These experiments at low temperatures necessitated constant attention; it was impossible to leave them unwatched for more than 10 minutes: as, unless the amount

of ice in the bath was very carefully regulated, the temperature was liable to change. For this reason I have at some of these temperatures contented myself with three readings instead of taking the usual four or five.

(iii) *Sources of Light.*

At the beginning of the investigation ordinary incandescent gas-light was employed. Ultimately this light was found insufficient, and the Keith high-pressure incandescent light was employed.

With the ordinary burners the gas pressure was kept constant at about 1 inch of water by means of a Moitessier regulator. Mantles of one make only were employed, either a new one for each experiment, or, where stated, one for a series of experiments.

The weakest light used, that obtained when the front of the mantle was 130 millims. from the leaf, is termed light of unit intensity.* By varying the distance between the leaf and the burner according to the law of inverse squares other intensities were obtained. A limit was imposed on the increase in the light which could be thus obtained, by the necessity of leaving a certain amount of space between the front glass of the leaf chamber and the window of the bath to insure free circulation of the water and consequent uniformity in the temperature. The greatest amount of light obtainable in practice from the single incandescent burner by increasing its proximity to the leaf was a light of sixfold intensity.

A somewhat increased intensity of light was obtained by using *two* burners side by side, but these could not be placed nearer to the window of the bath than 31 millims. (for fear of cracking it). The distance from the leaf was now 55 millims., and the intensity thus obtained was eightfold.

In the Keith burner the gas is supplied by a self-regulating water-motor under a very constant high pressure (8 inches of water), and this gives an extremely steady light.

To obtain a very intense light from this apparatus, three burners, arranged on a triangular plan, were employed. Two were placed side by side, in a plane parallel to that of the window of the bath and the third just behind, filling the small gap between them. These three burners were mounted on a brass stand, so that their relative positions remained unchanged, and they could be used as a single source of light.

The light of the three burners was compared, by means of a grease-spot photometer,

---

* All the intensities of light subsequently used are expressed in terms of this unit intensity by making due allowance for alterations in distance and differences in the burners employed. Of course with a compound source of light such as an incandescent mantle, such a comparison of intensities makes no pretence to accuracy, but may serve as a rough guide to the relative amounts of light necessary for maximal assimilation at various temperatures, and is absolutely necessary for convenience of reference.

It will soon also be made clear that knowledge of the exact intensity of light being used is not of critical importance in these investigations.

with a single ordinary incandescent burner, and it was found to be about six times that obtained from the single burner.

This relation has been adopted as the basis for comparing the intensities of the light, obtained in the later experiments, with those of the earlier ones.

These burners, although so excellent a source of light produce, however, a very large amount of heat. The simple system of circulating water employed before was quite inadequate to keep down the temperature of the bath. It was necessary to absorb more effectually the heat radiations before they reached the bath, by means of a special water-screen.

The screen employed consisted of an erect glass plate down both surfaces of which a thin sheet of water flowed rapidly. This was placed a few millimetres away from the window of the bath and the burners could be brought so close to the screen that only 24 millims. intervened between them and the window. With this arrangement the temperature of the bath itself could be kept constant at as low a temperature as 11° C.

With the ordinary incandescent burner the heat radiation was not sufficient to produce in the leaf, by reason of its greater absorptive power for radiation than that of water, any appreciable rise of temperature over the temperature of the bath. This was proved by wrapping the bulb of a delicate thermometer up in a piece of leaf, placing it at the proper distance from the light and comparing its readings with those of a similar thermometer with uncovered bulb. The excess temperature of the wrapped-up thermometer was not more than one-tenth of a degree even with the bath at the low temperature of −6° C.

With the powerful Keith burners, however, the effect was very different and the wrapped-up thermometer showed an excess temperature of 6° or 7° (tested with the bath at 11° C. and at 32° C.).

It was, therefore, quite obvious that here the temperature of the bath afforded no clue to the true temperature of the leaf, and that in order to obtain the true relation between assimilation and temperature when employing an intense light, it would be necessary to *determine the internal temperature of the leaf during each assimilation experiment.*

This, although introducing great additional complexity into the inquiry, was accomplished by employing thermo-electrical methods. A full description of these is given later. They showed that the temperature of the leaf, when exposed to powerful radiation may be 10° or more above the temperature of the bath.

(iv) *Procedure.*

Very suitable material for these investigations was yielded by the leaves of cherry

laurel, *Prunus Laurocerasus*, garden variety *rotundifolia*. To insure similarity in environment the leaves were always cut from the same side of one particular bush. Great care was taken to select them as far as possible of the same size and development, in order to reduce individual variation to a minimum.

Stress has already been laid on the importance of keeping these leaves under exactly similar conditions for some time before use. That the realisation of this ideal is attended with a certain amount of difficulty, the following considerations will show.

Since the effect of keeping isolated leaves in the dark, is to produce a continuous fall in the respiration, the amount of CO_2 given off by any leaf would vary with the number of days that it had been picked. On the other hand any increase in nutrition, *i.e.*, exposure of the leaf to a bright light, causes a decided increase in the respiration.

It was also impossible to use leaves straight from the tree; for it had been found (unpublished investigations by Dr. BLACKMAN and myself) that the respiration of a leaf during the first 24 hours after isolation might be extremely irregular. It was, therefore, necessary to wait at least 24 hours after cutting, before using the leaf, and yet to prevent either a diminution or increase in the respiration.

These demands were fulfilled by keeping the leaves with their stalks in water in covered beakers, exposed to diffuse light. Presumably they were just able to assimilate sufficiently to cover the loss in reserve material due to respiration. A fairly constant temperature was maintained, averaging between 10° C. and 14° C. at different times.

Good evidence that by this arrangement leaves could be kept in a uniform condition for several days, will be found among the records of the respiration experiments (Table I.). For instance, there are two determinations of the respiration at 14° C. These were both performed on the same set of leaves, which in the interval (a week) had been kept under the above-mentioned conditions. The agreement between the two sets of readings is most satisfactory.

The two determinations of the respiration at 33° C. were taken from two different sets of leaves, kept for varying lengths of time in the same way. Here also there is practically no difference in the readings.

In the assimilation experiments a fresh leaf was employed for each experiment. This, as mentioned before, constitutes one of the fundamental points of difference between this investigation and those which preceded it.

To avoid any loss of water-content by excessive transpiration during the experiment, the leaf stalk, with its surface freshly cut, was placed so as to dip under the water at the lowest point of the chamber. A comparison of the weights of the leaves before and after the experiment shows that this precaution was quite sufficient.

The general procedure of an experiment was as follows—A leaf (chosen from those kept under the above-mentioned conditions) was weighed and the area measured by means of a polar planimeter. It was then sealed up in the chamber and the latter was placed in the bath, which was already at the required temperature. The distance

between the source of light and the chamber was adjusted, the aspirators, clockwork CO_2 generator, &c., started and adjusted.

The currents were allowed to run for $1\frac{1}{2}$–2 hours before actual readings were taken, to render the conditions, such as CO_2 content of the chamber, &c., constant. Consecutive 2-hour readings were then instituted, and at the end of the experiment the leaf was removed, weighed, and any change in the appearance noted.

It may be of interest to give here two experiments which illustrate well the need for uniformity in the condition of the leaves before experiment. In both the cases given below the leaves were picked during a sharp frost and were used immediately for assimilation experiments at temperatures of 14° C. and 18° C. It will be seen that the value obtained for the assimilation rises throughout the experiment. This may be attributed to the gradual opening of the stomata from the winter closure, under the influence of the light and warmth, or, perhaps, partly also to the slow recovery from "assimilatory inhibition," that had been brought on by the continued frost, see EWART (16). In either case the effect lasted for 10 or 12 hours. Subsequent experiments on leaves, that had not been exposed to frost, showed that the highest number obtained represents the normal assimilation.

These experiments also form a good illustration of the value of the system of taking continuous readings. Any one of the readings singly would have been very misleading.

EXPERIMENT III.—December 17–18.   14°·2 C.

Weight before and after experiment. Area.	Temperature.		Time.	All amounts are expressed in grammes of CO_2.		
	Time.	Temp.		CO_2 supplied. Control tubes.	CO_2 unassimilated. Tubes from chamber.	Apparent assimilation. Difference.
1st weight, 1·38 grms.	9.30 P.M.	14·3	9–11 P.M.	Preliminary	Preliminary	—
2nd weight, 1·40 „	12.15 A.M.	14·2	11– 1 A.M.	·0297$\frac{1}{2}$	·0294	0003$\frac{1}{2}$
			1– 3 „	·0297$\frac{1}{2}$	·0277	·0020$\frac{1}{2}$
Area, 47·0 sq. centims.			3– 5 „	·0301	·0269$\frac{1}{2}$	·0031$\frac{1}{2}$
			5– 7 „	·0300$\frac{1}{2}$	·0266$\frac{1}{2}$	·0034
	9.30 A.M.	14·2	7– 9 „	·0297$\frac{1}{2}$	·0256	·0041$\frac{1}{2}$

EXPERIMENT IV.—December 9–10.   18° C.

Weight.	Temperature.		Time.	All amounts are expressed in grammes of CO_2.		
				CO_2 supplied.	CO_2 unassimilated.	Apparent assimilation.
	Time.	Temp.		Control tubes.	Tubes from chamber.	Difference.
Weight, 1·68 grammes	9 P.M.	18·1	9–11 P.M.	Preliminary	Preliminary	—
			11– 1 A.M.	·0320	·0284	·0036
			1– 3 ,,	·0322	·0285	·0037
			3– 5 ,,	·0325	·0277$\frac{1}{2}$	·0047$\frac{1}{2}$
			5– 7 ,,	·0319$\frac{1}{2}$	·0271	·0048$\frac{1}{2}$
	9 A.M.	18·2	7– 9 ,,	·0322	·0270$\frac{1}{2}$	·0051$\frac{1}{2}$

Section III.—EXPERIMENTS AT LOW AND MEDIUM TEMPERATURES.

(i) *Respiration.*

Since the determination of the real assimilation of a leaf necessitates a knowledge of the respiration at the time, it was important to determine this under conditions exactly similar to those to which the leaves were subjected in the assimilation experiments. The respiration at the temperatures considered in this section is small in comparison with the assimilation, and no critical work in this regard is required. At all temperatures below 0° C. the respiration could be neglected, for it was clear from the determinations at 5° C. that below 0° C. the amount for a single leaf would come within the experimental errors.

The procedure adopted in determining the respiration at medium temperatures was similar to that employed in an assimilation experiment, with the exception that the two currents were in this case freed from CO_2, and an estimation was made of the amount of CO_2 added to one of them in a given time by the plant. Since the respiration of a single leaf was very small, several leaves were taken for each determination, the number of leaves being regulated to give an amount of CO_2 convenient for measurement.

The results obtained in these determinations of the respiration are given in Table I. The consecutive readings in any one experiment show good agreement, so do also the duplicate experiments. Moreover, when the respiration of a single leaf is calculated from these results, small inaccuracies in the actual determinations are very much reduced. Even the slight fall in respiration, which normally occurs during a prolonged experiment in the dark, is practically eliminated in the reduced numbers.

A curve showing the respiration at different temperatures is given in fig. 2, p. 65. The numbers employed in its construction are the average values obtained from each experiment. Here the respiration, in all cases, has been calculated for a constant weight of 2 grammes.

The assimilation results are calculated in all cases for a leaf surface of 50 sq. centims. The weight of this is approximately 2 grammes, so that the respiration and assimilation results are more or less comparable.

(ii) *Experiments with Unit Intensity of Light.*

Full details of the experiments to be considered under this head are given in Tables II., III. and IV. at the end of the paper. The various columns explain themselves, and but few additional remarks are needed.

The temperature indicated by the thermometer in contact with the leaf chamber was noted at intervals throughout the experiment. These readings, with the time at which they were taken, are given in the second and third columns. In the low temperature determinations, where the experiment was watched the whole time and the temperature regulated by the amount of ice added, this seemed superfluous, and more general statements are given.

The eighth column shows the respiration (deduced from the results given in Table I.) for the weight of the individual leaf used in the assimilation experiment. Since all the cells take part in the respiration, it seemed more correct to refer the respiration to the weight of the leaf than to the area of leaf surface as was done by KREUSLER (1). The assimilation, on the other hand, varies with the leaf surface.*

The only column which contains numbers directly comparable with those obtained in other experiments is the last. Here allowance has been made, not only for the respiration, but also for the leaf area. The assimilation for 50 sq. centims. and 1 hour has been taken as the standard.

Table II. gives the results of assimilation experiments performed between the temperatures 14° C. and 33° C.

Similar experiments are described in Table III., but here the temperature limits are 5° C. and 25° C. The results of experiments at temperatures between − 6° C. and +8°·6 C. are given in Table IV. As mentioned before, the respiration below 0° C. could be neglected. A slight difference in the procedure was necessarily adopted, and no water was placed in the leaf chamber in any of the experiments at temperatures below 0°. C.†

---

* Where the respiration at a particular temperature has not been found experimentally, the amount has been calculated from the respiration curve given in fig. 2.

† The following experiment shows that this lack of water had no inhibitory effect (at low temperatures) on the assimilation.

A leaf was set up in the chamber in the usual way, but without water. Three readings of the assimila-

The detailed results given in these tables are represented in the curve (fig. 2). The latter embodies the mean result of every experiment. Each point represents the average of the readings obtained in a single experiment only.

Although, without exception, every experiment made is represented in fig. 2, yet there are only two numbers which are anomalous, namely, two experiments at 14° C. (located by error at 12°·5 in the figure) and of these the mean falls perfectly into line.

Fig. 2.
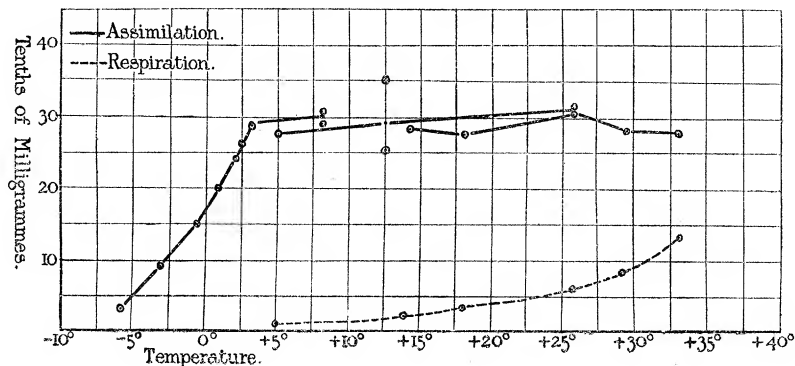
tion were taken at the temperature of 5°·8 C. Water was then introduced into the chamber, and, after allowing the conditions to become constant, readings were again taken at the same temperature. The result was conclusive; no rise at all was produced in the assimilation, rather there was the inevitable falling off, due to the length of the experiment.

EXPERIMENT XXV.—February 18. 5°·8 C.

| Weight and area. | Temperature. | | Time. | All amounts are expressed in grammes of CO_2. | | |
	Time.	Temp.		CO_2 supplied. Control tubes.	CO_2 unassimilated. Tubes from chamber.	Apparent assimilation. Difference.
1st weight, 1·57 grms.	10.45 A.M.	5·6	10.15–11.45 A.M.	Preliminary	Preliminary	—
	11.45 ,,	5·7	11.45– 1.45 P.M.	·0309	·0256½	·0052½
Area, 43·0 sq. centims.	2.30 P.M.	5·7	1.46– 3.45 ,,	·0311½	·0264	·0047½
	3.38 ,,	5·8	3.45– 5.45 ,,	·0311½	·0271½	·0040
			Water added			
			5.45– 6.36 P.M.	Preliminary	Preliminary	—
	6.45 ,,	6·0	6.36– 8.36 ,,	·0315½	·0275½	·0040
			8.36–10.36 ,,	·0313	·0272½	·0040½
	12.10 A.M.	6·1	10.36–12.36 A.M.	·0310	·0273½	·0036½

The curve is made up of three parts, denoting the result of the three series of experiments given in Tables IV., III. and II. respectively. The temperature range of each series has been so chosen as to overlap that of the next. This arrangement enables a direct comparison to be made between them, and gives confirmatory evidence for part of the temperature range.

The slight dislocation between the parts where they overlap is most probably an expression of the difference between the light produced by the two incandescent mantles employed. Thus this amount is shown to be not of sufficient magnitude to cause any effective disturbance of the curve.

The first point to be noticed is that assimilation can be detected as low as $-6°$ C. Previous workers with less exact methods have not detected it below $0°$ C.[*] e.g., CLOËZ and GRATIOLET (6), 10–15° C. for *Potamogeton*, &c., HENRICH (10) 2–4° R. for *Hottonia*, and BOUSSINGAULT (7) $+0.5$ to $+3°$ C. for *Pinus laricio* and grasses. This is due partly to the methods of observation, and in some cases to the choice of plants. Delicate water-plants would most probably cease to assimilate long before the hardy evergreen cherry laurel. It is very probable that cold-loving plants, such as conifers, lichens, &c., may assimilate at even lower temperatures.

The next point to be noticed is the sharp change in the character of the curve at $+3°$ C. There is an extraordinary uniformity in the assimilation from $+3°$ C. to 33° C. Individual differences in the readings of any one series are hardly greater than the experimental error obtained in the actual experiments, and this, considering that separate leaves were used each time, and that slight variations in the light, &c., were possible, is most satisfactory and completely demonstrates the validity of the methods adopted in the treatment of the leaves.

Below $+3°$ C. we find the assimilation suddenly decreasing very rapidly with the temperature, until, at $-6°$ C., the leaf has practically lost this faculty. This curve of decrease of the assimilation is well established, for the intervals of temperature between the successive determinations are extremely small. There are no less than seven separate determinations in the range from $-6°$ C. to $+3°$ C. Since the assimilation was changing so rapidly, a large number of experiments at slightly different points, seemed more conclusive than several determinations at one or two, more widely separate, temperatures.

This marked contrast between the nature of the assimilation curve above and below 3° C. is so remarkable that it suggested some influence other than the direct effect of the temperature. On reviewing the factors influencing assimilation which might possibly be affected by a low temperature, there seemed but one probable one, partial closure of the stomata. This might well be brought about by intense cold, resulting in a very reduced value for the assimilation. A separate experiment was, therefore, undertaken to test the validity of this hypothesis.

---

[*] As has been shown in the Introduction, it is difficult to accept JUMELLE'S results at $-30°$ C. and $-40°$ C.

If artificial openings be made in a leaf with closed stomata a great increase in the assimilation results. The assimilation of a certain leaf was taken at $-3°·6$ C. After two readings the leaf was removed and cut in 40 places pinnately with a sharp scalpel. The leaf was then replaced in the chamber and readings were again taken. Had the

EXPERIMENT XXVI.—February 24 and 25, 1902. $-3°·6$ C.

Weight and area.	Temperature.	Time.	All amounts are expressed in grammes of CO_2.		
			CO_2 supplied.	CO_2 unassimilated.	Apparent assimilation per 2 hours.
			Control tubes.	Tubes from chamber.	Difference.
Weight, 1·78 grms. Area, 48·7 sq. cms.	$-3°·6$ C. to $-3°·8$ C.	10.15–11.45 A.M. 11.45– 1.45 P.M. 1.45– 3.45 ,, 3.45– 5.45 ,,	Preliminary ·0303 ·0300½ ·0312½	Preliminary ·0282 ·0283½ ·0297	— ·0021 ·0017 ·0015½

Leaf in chamber in light during night; temperature about 7° C. At 11 A.M., cut 40 cuts pinnately.

	$-3°·6$ C.	11.15–12.45 A.M. 12.45– 4.55 ,, 4.55– 6.55 ,,	Preliminary ·0606 ·0288	Preliminary ·0587½ ·0277½	— ·0009 ·0010½

stomata been closed there must have resulted a great increase in the assimilation. This was not the case; in fact there was a decrease owing to the duration of the experiment. The rise in the respiration resulting from the wounding of the leaf could introduce no error, since at so low a temperature the respiration even if doubled or trebled would still be negligible.

As a contrast to the above an experiment may be given in which cutting produced an increase in the assimilation. A leaf dried over calcium chloride assimilates very feebly. On cutting, the CO_2 absorption became very much greater, showing that the small amount obtained before was due to closure of the stomata (see Exp. XXVII.).

As regards the change in form of the curve (fig. 2), Experiment XIX., Table IV., in which the temperature was gradually raised during a reading, gives a very good demonstration of the fact that at $+3°·6$ C. the value of the assimilation is already quite the same as that represented by the level portion of the curve, while numerous other experiments show that this is not the case for the values below $+2°$ C.

It was impossible to believe that the influence of the temperature on the assimilation really changed its character in the sharp and sudden manner the curve suggested, and yet the true interpretation was not at first sight obvious.

EXPERIMENT XXVII.—February 21.

Weight and area.	Temperature.		Time.	All amounts are expressed in grammes of CO_2.		
				CO_2 supplied.	CO_2 unassimilated.	Apparent assimilation.
	Time.	Temp.		Control tubes.	Tubes from chamber.	Difference.
Original weight, 1·98 grammes; dried over CaCl₂, weight, 1·75 grammes	3.45 P.M.	9°·2	2.50– 4.50 P.M.	Preliminary ·0305½	Preliminary ·0301½	— ·0004
			4.50– 6.50 ,,	·0314	·0309½	·0004½
			6.50– 8.50 ,,	·0306½	·0304½	·0002
	9 ,,	8·0	8.50–10.50 ,,			
			Cut at 11.30 ,,			
Area, 53·1 sq. centims.	11 ,,	8·2	—	—	—	—
	1 A.M.	8·2	12.6 – 2.8 A.M.	Preliminary ·0310	Preliminary ·0259	— ·0051
			2.8 – 4.8 ,,	·0310	·0259	·0051
			4.8 – 6.8 ,,	·0307½	·0254½	·0053
	11.15 ,,	7·6	6.8 – 8.8 ,,	·0310	·0252	·0058

A very valuable suggestion was then made to me by Dr. BLACKMAN, which provided the key to the whole problem.

He suggested that only the first sloping part of the curve corresponded to a condition of things in which the amount of assimilation was really a direct function of the temperature, and that the level second part was simply an expression of the full amount of CO_2 that the energy of the light provided was able to decompose. This amount is governed by thermo-chemical considerations, and is not increased by raising the temperature.

The limiting assimilation maximum, fixed directly by any given temperature, can then only be arrived at when the light is adequate for the decomposition of the amount of CO_2 in question, and when, also, of course, that amount of CO_2 is freely available. Therefore, to reach these maxima for the higher temperatures, more light must be employed, and the evidence that is to be looked for to show that the limit is reached, and that the light really is sufficient, will be of the nature of showing that, at the given temperature, increase of light no longer augments the assimilation.

These considerations are fully borne out by experiment, and it is found that, measuring the assimilation over the whole temperature range in a low intensity of light, gives a curve (as in fig. 2), with its changing point, at a low temperature; working with a somewhat higher intensity, gives the change at a higher temperature (as in fig. 3), while, if the light is very intense, the change to the level portion comes very high up, or, indeed, not at all (fig. 5), the curve being instead

interrupted by the effect of sub-fatal temperature. At high temperatures, then, it is only with such intense light that the pure temperature effect can be evaluated.

It is interesting to note that the final curve put forward by KREUSLER is really of the imperfect compound nature indicated. His procedure, however, was not perfect enough to bring out the changing point as a sharply-defined effect. It looks in his curve merely like a slow change in steepness, such as might be a direct temperature effect. The more accurate methods here employed brought out the changing point as so sudden and abrupt an effect that it could not be overlooked.

(iii) *Experiments with Various Low Intensities of Light.*

These experiments were undertaken to prove the truth of the hypothesis just brought forward.

At three different temperatures—$+0°·4$ C., $+9°$ C., $+11°$ C.—the assimilation with the unit intensity of the light (*i.e.*, that used in the previous experiments) was determined and then the light was increased until no further increase was produced in the assimilation. A different limit for the assimilation was found in each case, and, to reach it, a greater intensity of light was necessary the higher the temperature. In the experiments at 25° C. this limit was not reached even with a sixfold intensity of light.

Table V. embodies the results of these experiments. The arrangement is, in the main, the same as that in the preceding tables. Column 1 contains a statement of the relative intensities of the light. These were obtained by shifting the leaf chamber in relation to the window of the bath, while the position of the burner was unchanged.

Notifications of the temperature in some of the experiments in Table V. are very few, because water running rapidly through the bath was used, and this gives a very constant temperature.

Readings with the leaf chamber in the "unit light" position were first taken, then the chamber was moved into the position required for the greater intensity and determinations were again made, after a short preliminary to obtain equilibrium of conditions. It was found impracticable to determine the effect of more than two different intensities of light on the same leaf.

Table VI. gives a summary of these results. Here both the apparent and the real assimilation* for 50 sq. centims. and 1 hour are given, being the average of all the individual results of Table V., also the ratios between the amounts of the real assimilation at double, fourfold and sixfold intensities, to that obtained with light of unit intensity.

---

* The real assimilation has been obtained from the apparent in the same way as before by using the results given in Table I.

If we now consider the results under the heading of " Real Assimilation," we find that the amount of assimilation obtained with light of unit intensity is approximately constant* at all temperatures except the lowest. Here, as we should expect from the previous results, the amount is lower. Not even all the amount of light represented by L.In. = 1 can at the low temperature be utilised.

When the intensity is doubled, the assimilation at $0°·4$ C. is unaltered, but an increase is produced at the other temperatures, numbers almost identical being obtained for all of these. When four times the light is used we find a similar phenomenon. The assimilation at $9°$ C. is no greater than that corresponding to twice the light, for the temperature exerts its limiting effect and the leaf can assimilate no more. At $11°$ C. a higher number is obtained, which, however, is not so great as that obtained at $25°$ C., showing that the leaf can make use of more light than is given by L.In. = 2, but it cannot use all that of L.In. = 4. Sixfold light gives practically the same result as fourfold light for $11°$ C., showing that the maximum has been attained.

Thus at the temperatures of $0°·4$ C., $9°$ C. and $11°$ C., by sufficiently increasing the light, a limiting amount of assimilation has been obtained, different in each case.

Fig. 3.
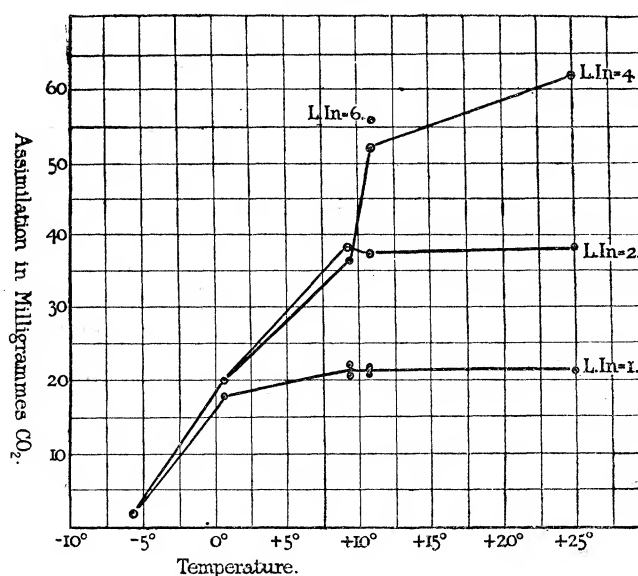
Fig. 3 represents graphically the results of Table VI. Three different curves are obtained, one with the unit intensity of light, one with twice the light, and one, the true assimilatory curve for the temperatures from $-6°$ C. to $+11°$ C., with fourfold light. The truth of the hypothesis is, therefore, well-established.

* Experiment XXXVI. must be considered merely as regards the ratio yielded. The results obtained for L.In. = 1 and L.In. = 4, though harmonious, are far too small in comparison with the other experiments, indicating that the leaf was probably in an abnormal condition.

In considering the ratios given in the last four columns, it is of interest to note that, although the assimilation is not maximal, light of double the intensity does not produce fully double the assimilation. It would seem as if, a little below the limit, the leaf could not respond completely to the light.

Thus the law that the assimilation varies with the intensity of the light appears only to be true for degrees of illumination well removed from that necessary to produce the maximal amount. This needs further investigation, but 'some determinations by REINKE (17), are, to a certain extent, confirmatory. He found that, after a certain limit, increase in the intensity of the light produced no effect on the assimilation, and that, just below this limit, the increase in the assimilation was not proportional to that of the light.*

Section IV.—EXPERIMENTS AT HIGH TEMPERATURES.

(i) *Respiration.*

Before dealing with the actual assimilation numbers obtained at these temperatures it will be necessary to again consider shortly the question of the respiration. The higher the temperature the more important a knowledge of the true value of this function becomes.

Unfortunately the question of the behaviour and value of the respiration during exposure to high or even to comparatively high temperatures is very complicated.

A separate inquiry was made into the general behaviour of the respiration of leaves under various conditions at high temperatures. Many interesting results were obtained, having an important bearing on our whole conception of the function of respiration and its reaction to changes in condition. A full account of this investigation will be published separately, but it will be necessary to state here very shortly some of the results that bear on the present question.

The value of the respiration changes rapidly with the time of exposure, and consecutive 2-hour estimations of the respiration of the single leaf, prove that, instead of forming a smooth falling curve, the CO_2 produced by the single leaf shows a large number of oscillations, much too great to be due to experimental errors. Moreover, the individual variation in the respiration of the leaves under similar external conditions is considerable, due, no doubt, in great measure, to differences in internal condition, such as amount of carbohydrates present, &c. Such individual variations and oscillations occur at the low temperatures also, but are so small in amount that they are not greater than the experimental errors. It is, therefore, quite impossible to predict with certainty the value of the respiration of a leaf at any particular moment by observations on another leaf under the same external conditions.

* It will have been observed that the assimilation obtained with unit light is much lower than that obtained in the earlier experiments with the same light. This is due to a seasonal change in the condition of the leaves on the tree, and will be dealt with later, see pp. 82 and 83.

However, these factors, although exerting an appreciable influence, are nevertheless insignificant when compared with the enormous effect produced on the respiration by assimilation. *The respiration is continually changing during an assimilation experiment by virtue of the assimilation*, and may be doubled by an exposure of 4 or 5 hours to light and CO_2. This is due doubtless to the production of a fresh supply of carbohydrates, but the rise produced is not entirely proportional to the amount of CO_2 decomposed.

Thus as it is at present impossible to determine directly respiration in presence of assimilation, we have no means of obtaining the exact value of this quantity, and we are obliged to make an approximation.

It will be obvious that, if the respiration of the leaf be taken in the dark, before the assimilation experiment, too low a value is obtained; if taken after the assimilation, the value tends to be too high. By taking the mean of these two determinations, we must, however, obtain an average value which cannot be very different from the real value during the assimilation, and is certainly accurate enough for the purposes of these experiments.

This is the plan that has been adopted in the experiments at high temperatures given in Table VII. In the experiments of Table XI. it was impracticable to determine the respiration twice owing to the complexity of the experiment and the necessity of completing this series of experiments within a short time.* Under these circumstances the respiration was taken *after* the assimilation only. The value employed for the respiration is, therefore, admittedly too high, but the difference from either the average value or the true value is not sufficient to seriously affect the values of the real assimilation nor to invalidate the form of the curve.

(ii) *Experiments with Eight-fold Intensitiy of Light.*

In these experiments the source of light consisted of two ordinary incandescent burners placed side by side at a distance of 55 millims. from the leaf, giving a light of eight times unit intensity.

The leaf was set up in the usual manner, and, after an hour's preliminary, two 2-hour estimations of the respiration were made in the dark. The leaf was then exposed to the light, and several 2-hour estimations of the apparent assimilation taken. The chamber was then darkened and the respiration again determined.

In order to reduce as much as possible the necessary preliminary between exposure to light and darkness, the current containing CO_2 was drawn through the apparatus during the whole time, and thus, a departure from previous procedure, the respiration was estimated as an addition to the current of CO_2 passing through the chamber.

Determinations of the assimilation were made at temperatures of 11° C., 25°·4 C.,

---

* The condition of the leaves was rapidly changing owing to a change in the season, see p. 82.

32°·1 C., 38°·3 C., 40°·9 C., and 42°·9 C. (Table VII., fig. 4). The results of the two experiments made at each temperature are in good agreement.

In Experiment XXXVII. the respiration was not estimated, but was calculated as before, from Table I., it being impossible to determine accurately, with the apparatus employed, the CO_2 produced at 11° C. by a single leaf in 2 hours.

In this experiment after three 2-hour determinations of the assimilation with the light of eight times unit intensity, the light was changed to the single incandescent burner at a distance of 42·5 millims., giving an intensity of light represented by 4·6 times unit intensity. No change, however, was produced in the amount of the assimilation, showing that the lesser intensity of light is sufficient for maximal assimilation at this temperature.

That this is so may also be shown by reference to the other experiments in the series. At 11° C. the amount of assimilation obtained was ·0068 gramme per hour. By raising the temperature to 25° C., the value of the assimilation was increased to ·0126 gramme per hour with no alteration in the light provided. It is clear, therefore, that, since the light was sufficient to decompose ·0126 gramme of CO_2 per hour, the small amount of assimilation obtained at 11° C. was not due to insufficiency of illumination. Since the leaf has therefore more than enough light and CO_2, the value obtained for the assimilation must represent the maximal assimilation of the leaf at 11° C. It is impossible to obtain a greater assimilation except by raising the temperature.

Let us consider now the case of the other experiments. Since the value obtained throughout the range of temperature 25–38° C. is the same, it is clear that this is an expression of the amount of CO_2 which can be decomposed by the light supplied, and bears no relation to the temperature. The leaf assimilates as much as the illumination provided allows, if it is not limited by the temperature it is so by insufficiency of the light.

At temperatures higher than 38° C. the assimilation falls much below the limit imposed by the illumination, showing that here the amount is limited by some condition other than that of the light. Rise in temperature has begun to exert an injurious effect. At 45° C. the influence of the temperature alone is sufficient to kill the leaf. It is not surprising that, a few degrees before this point is reached, the temperature, though not fatal, should yet be harmful and produce a diminution in the amount of assimilation of which the leaf is capable, such diminution being greater the nearer the temperature to the temperature of death.

From a consideration of the numbers obtained in these experiments we should expect that it would be impossible to produce greater assimilation at 41° C., 43° C., but that under the influence of increased light the leaf should be capable of greater assimilation at temperatures between 25° C. and 38° C. ; possibly at these temperatures themselves.

It will be as well to consider here the evidence that when employing two ordinary

Fig. 4.
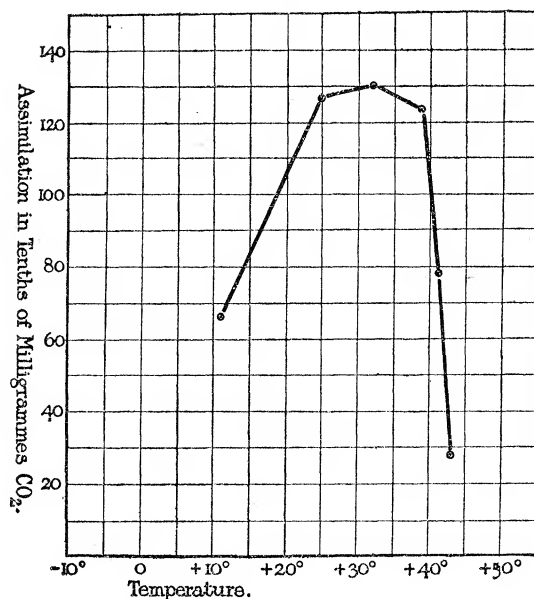
incandescent burners as the source of light, the temperature of the leaf was not appreciably different from that of the bath. In the experiment at 11° C., the light was changed considerably in intensity without producing an alteration in the amount of the assimilation. If the light had raised the temperature of the leaf above that of the bath, the substitution of the less intense light must have lowered the leaf-temperature, and therefore made a difference in the possible amount of assimilation. This is good evidence that at 11° C. the leaf was not appreciably affected in temperature by this light and thus, if this were so at 11° C. when there was excess of light, it would also be so at the other temperature when the light was obviously insufficient.

(iii) *Experiments with High Intensities of Light.*

(a) *Preliminary Experiments.*

The results of the first of experiments performed with the Keith high-pressure burners are given in Tables VIII., IX., and X. Those in VIII. and IX. were carried out before the necessity of determining the internal temperature was realised. In these, and all succeeding experiments, it was necessary to supply a greatly increased amount of CO_2, in order that the results might in no way depend on the percentage of CO_2. In consequence, in order not to risk incomplete absorption in the Pettenkofer tubes, each was used for only 1 hour. Since the amounts of CO_2 to be determined were so large, there was no objection to using this shorter time interval. The true temperature of the leaf was not determined in these preliminary experiments, but must have been several degrees higher than that of the bath. This

is clearly shown by the fact that the value obtained for the assimilation when the bath was at 11° C., is considerably higher than the amount shown in Section IV. (ii) to be maximal for that temperature.

The highest value for the assimilation we have found occurs in Experiment L. I believe that this is also the highest value recorded by any observer.

Table IX. gives the results of some preliminary experiments illustrating the rise in the temperature of the leaf produced by the direct radiation of an intense light. The temperature of the bath was 32° C., a temperature at which a leaf can live for over a week without being outwardly affected. Under the influence of the intense light, however, large brown patches appeared, the leaf became flaccid, and lost very rapidly a third of its weight, its behaviour thus resembling that of a leaf which has been exposed to a temperature of 43° C. or 45° C. The values obtained for the assimilation are very low, and it will be remembered that in Section IV. (ii), the experiments with weak light, it was shown that there was rapid decrease in the assimilation at temperatures above 38° C.

An interesting experiment is recorded in Table X. The assimilation of one leaf was determined under varying intensities of light, the temperature of the bath being 11° C. The arrangement of three Keith high-pressure burners was employed, the intensity of the light being altered by increasing their distance from the leaf. Now had the leaf been at the same temperature as the bath, the highest intensity of the light should have given a value of the assimilation of ·0066 gramme per hour approximately (according to Experiment XXXVII.), and no change should have been produced in this value until the light had been decreased to an intensity less than fourfold. Table X. shows that the actual results obtained were very different. The initial value of the assimilation was ·0100 gramme per hour, and this decreased continuously. There was no indication of the existence of a maximal amount of assimilation. This is entirely due to the heating effect of the excess light. Each additional increase in the intensity of the light produced a further increase in the temperature of the leaf, and, therefore, permitted of an increase in the assimilation.*

These preliminary experiments afford conclusive evidence of *the necessity of determining the internal temperature of the leaf during an assimilation experiment when using an intense light.*

(b) *Method of Determining the Internal Temperature of a Leaf.*

The temperature was determined by measuring, by means of a galvanometer, the electromotive force produced by the difference in temperature of two thermo-elements, one being embedded in the tissue of the leaf and the other being kept at a known temperature.

* The probable temperatures were determined later electrically by placing another leaf in exactly the same positions. The results obtained will be found in Column 3 of Table X.

The galvanometer used was an ordinary Thomson mirror-galvanometer, about $\frac{1}{2}$ ohm in resistance. The deflections were read directly on the scale ; no refinements, such as the use of a telescope, were employed. A deflection of about five scale-divisions corresponded to a change in temperature of one degree, and it was possible to read to half a division. The error in the temperature determinations was found to be about half a degree, so that the direct method of reading the deflections was well within the limits of accuracy imposed by other conditions.

The thermo-elements were composed of copper and constantan (an alloy of copper and nickel), as these give a relatively large electromotive force.

It was a matter of some difficulty to obtain a thermo-element sufficiently small to be inserted in the leaf. Satisfactory junctions were made for this purpose by the Camb. Sci. Inst. Co. Extremely fine wires of copper and constantan were joined so skilfully that the junction was not much thicker than the wires themselves, and this was afterwards filed down until the place of union was hardly distinguishable from the general substance of the wire. Naturally they were in consequence extremely fragile and difficult of manipulation,* for the wires themselves were so fine that they frequently broke if sharply bent. It was a difficult and tedious task to embed these elements in the lamina, and since experiment showed that the temperature registered was the same in both positions, they were always embedded in the tissue of the midrib. The latter was first pierced, parallel to the blade of the leaf, with a very fine needle, the end of the wire was pushed through this tiny canal by forceps, and the thermo-element was then drawn through until the junction was just buried in the midrib. To avoid any chemical action of the leaf-juices on the wires, and to prevent any possible short circuiting through the tissue of the leaf, the element was insulated by coating it with a very thin layer of varnish, flexible enough not to crack when bent.

A specialised form of leaf chamber was necessary which should permit the wires from the thermo-element to pass out for connection with the galvanometer. An adaptation was made in the back of the chamber ordinarily used. A cork bored with two holes was fitted into the centre of the glass. Two narrow glass tubes, U-shaped, passed through the cork and formed an appendage to the chamber. A small quantity of mercury was poured into the bend of each tube,† and wires of copper and constantan, similar to those employed in making the thermo-element, were pushed down the long free limbs of these tubes until they dipped into the mercury, when they were fixed in position by inserting a small wooden peg into the open end of the glass tube. The free ends of these wires, *i.e.*, those not in the

---

* On account of their fragile nature it was necessary to use a new thermo-element for almost every determination. The diameter of the wires was only ·087 millim.

† In spite of the small surface of mercury exposed, it was found that enough vapour was given off to seriously affect the assimilating leaf, turning it black. This was due to the marked poisonous action of mercury vapour on green plants *when assimilating*, and was effectually prevented by covering the surface of the mercury with a film of water; see BOUSSINGAULT (18).

mercury, served to make connection with the other element and with the galvano-meter. The wires of the thermo-element, projecting from the leaf, were inserted into the short limbs of the U-tubes and were carefully pushed down by forceps until their ends also dipped into mercury, and were, therefore, in electrical connection with the wires already in the U-tubes.

The glass, with its appendage of U-tubes and with the leaf in position, was then waxed on to the chamber.

The following diagram of the chamber and one of the two U-tubes may, perhaps, make this arrangement clear.

This arrangement of the U-tubes with the mercury and the accessory wires was, of course, permanent. The setting up, required afresh for each experiment, consisted in thread-ing an element through the leaf, inserting the ends of these wires in the tubes, and waxing the glass on to the leaf chamber, the most difficult part of the manipulation being the insertion of the ends of the thermo-element into the U-tubes without breaking the wires. Suffi-cient water was afterwards introduced through the air-inlet tube to cover the petiole of the leaf. The leaf chamber was then placed in position in the bath and connected with the CO_2 current and absorption apparatus.

Section through leaf and chamber and one of the two U-tubes.

Another thermo-element at a known tem-perature was, of course, necessary. This second thermo-element (which, for convenience of reference, I will term the control-element) was threaded along an indiarubber tube of very narrow bore to insulate it, and to prevent contact with the water. This rubber tube was then tied to a thermometer, so that the thermo-electric junction was just against the bulb. This stood in a vessel of water which could be brought to any desired temperature.

Very thick copper wire was employed for making the connections with the galvano-meter, since the latter had to be placed in the next room to avoid disturbances due to the working of the water-motor supplying the light. It was found most expedient to connect these wires to those from the thermo-elements by means of mercury cups. These were all fixed on the edge of the bath to insure equality of temperature, lest any slight difference in purity between the thin and the thick copper wires might cause them to act as secondary thermo-elements. The constantan wire from the thermo-element in the leaf was connected with that from the control thermo-element by another mercury cup.

The temperature was not read directly by means of the galvanometer, but a zero

method was employed, *i.e.*, the temperature of the water round the control element was adjusted until there was no deflection. The temperature of the control thermometer then indicated the real temperature of the leaf. This method worked very satisfactorily, and it was easy to determine the temperature to within half a degree, an accuracy quite sufficient for the purpose.

(c) *Final Experiments.*

In Section III. experimental evidence was given showing that from $-6°$ C. to $+11°$ C., if sufficient light be provided, the assimilation increases with the temperature. In the present section it has been possible to prove, by determining the internal temperature of the leaf, that this relation is maintained to within a few degrees of the death-point of the leaf.

The general course of procedure was as follows:—Immediately after setting up, the leaf was exposed to the light, and after a preliminary of $1\frac{1}{2}$ hours, four estimations (1 hour each) of the assimilation were made. The internal temperature of the leaf was determined at the beginning and end at least of every reading. A current of air free from CO_2 was then substituted and the light removed. The leaf was now at the temperature of the bath, *i.e.*, several degrees below what it had been during exposure to the light, the temperature at which its respiration was required. Hot water was therefore poured into the bath until the bath-thermometer registered the same temperature as that found, by means of the thermo-element, for the leaf during assimilation. The bath-thermostat was readjusted and the bath darkened by a black cloth. After an interval of 2 hours, two or three 2-hour estimations were made of the respiration.

All the leaves used in this series were picked on the same day and kept in beakers in the manner described on p. 61.*

As we can no longer (on account of the rise in internal temperature produced in the leaf) prove that the assimilation is maximal by merely showing that an increase in light produces no effect, it will be necessary to discuss each experiment separately and to prove that in each individual case we are dealing with the maximal assimilation for the particular temperature.

The true temperature of the leaf is dependent partly on the temperature of the bath and partly on the intensity of the light. The greater the intensity of the light, the greater must be the difference between the temperature of the bath and the required temperature of the leaf.

Experiment LVI., 15° *C.*—The intensity of the light employed in this experiment was thirteen times unit intensity. The lowest temperature of the bath attainable was 11° C. (that of running water at that time of year), and, in consequence, the

---

* Since the leaves were just passing through a period of seasonal change (see p. 82) it was necessary, in order to insure uniformity of condition, to gather them all on the same day.

excess of light must not be great enough to raise the temperature of the leaf more than 4° C.

Fig. 5.
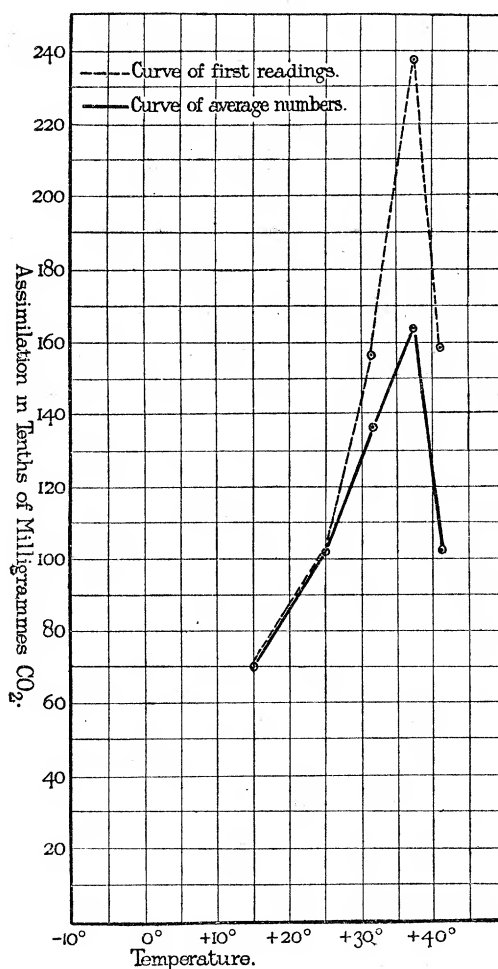
Under these conditions the leaf decomposed ·0070½ gramme CO_2 per hour. Now it will be seen that in Experiment XXXVII. at 11° C. the intensity of the light used was eight times unit intensity, *i.e.*, a little more than half that available in this experiment. The amount of CO_2 decomposed per hour was, however, ·0072 gramme, and therefore this leaf at 15° C. must be exposed to nearly twice the light necessary for the assimilation which it has actually performed. There is no doubt, therefore, that ·0070½ is the present true value of the maximal assimilation at 15° C., the excess of light being used up in raising the temperature of the leaf from 11–15° C., and in maintaining it at this temperature despite the loss by radiation to the bath.

It will be seen from the *précis* of this experiment given in Table XI. that only one long estimation of the respiration was made. The amount of CO_2 produced by a single leaf at 15° C. is so small that it is impossible to make 2-hour estimations with sufficient accuracy.

It should also be noticed that there is very good agreement between the successive

numbers obtained for the value of the apparent assimilation, proving that at this temperature there is no decrease in the assimilation due to prolonged exposure.

Experiment LVII., 23°·7 *C.*—The temperature of the bath was again 11° C., but the intensity of the light was twenty-six times unit intensity.

The value of the assimilation is ·0101 gramme per hour, less than the amount (·0128) induced in Experiment XXXVIII., Table VII., by one-third of the light. Here again, therefore, there is no doubt that we are dealing with the maximal assimilation for 23°·7 C.

At this temperature, also, there is no decrease in the apparent assimilation during the course of the experiment.

Experiment LVIII., 30°·5 *C.*—To obtain this temperature an intensity of light equivalent to L.In. = 45 and a bath temperature of 13°·5 C. were employed. To show that here the assimilation is maximal we must turn to the experiments in Table VIII. There the leaf chamber was in exactly the same position in the bath, the same burners were used, but were placed several millimetres further away. The value of the assimilation induced by this less intense light was, however, ·0249 gramme per hour, showing that the light employed in the present experiment would have been sufficient to produce more assimilation than this, and therefore the value of ·0136 gramme per hour must represent the maximal assimilation at this temperature.

Here the value found for the apparent assimilation decreases during the course of the experiment, the most rapid fall taking place between the first two readings. This is due mainly to the diminution in the assimilatory activity of the leaf when the absolute amount of assimilation is high.

Experiment LIX., 37°·5 *C.*—The intensity of the light used was the same as in the previous experiment, and, therefore, the same arguments apply, since the assimilation, even in the first reading, is not as great as ·0249 gramme per hour. Thus we have here again maximal assimilation.

In this experiment the sudden and large changes in the value of the apparent assimilation are very striking. The value obtained in the first reading is very large, that in the second is only two-thirds of the first. This fall continues during the third and fourth readings, but is less rapid. The average value obtained from the four readings, however, is quite in harmony, as regards its position in the curve, with the results obtained at the other temperatures.

Experiment LX., 40°·5 *C.*—The intensity of the light used was the same as in the last experiment, and since the assimilation is so much less, it must *a fortiori* represent the maximal value.

Here the high temperature exerts a decidedly injurious effect. Although the temperature is only 3° above that of the previous experiment, the value of the assimilation in the first readings has fallen from ·0237 to ·0149.

The temperature (37°·5 C.) forms the turning point in the curve. Two influences are

at work, firstly, the effect of the temperature hitherto experienced, *i.e.*, that of raising the amount of assimilation of which the leaf is capable, and, secondly, the injurious effect of a high temperature of which the tendency is to depress the assimilation. (See experiments at 41° C. and 43° C. in Table VII.)

This, the highest point of the curve, is, therefore, a position of unstable equilibrium. At the beginning of the experiment the leaf is under the optimal conditions for assimilation, and we get the greatest possible value for the maximal assimilation. Immediately after this effort, however, the injurious effect of the temperature gains the ascendancy, and we have the initial rapid fall* which subsequently becomes more gradual. That the optimal temperature for the assimilation must produce effects of this nature is obvious, for if the effect of rise in temperature is to become of a completely opposite character, there must be some point or region where both these tendencies make themselves felt.

At 15° C. and 23°·7 C. there was no change in the value of the assimilation during the experiment; at 30°·5 C. the first reading was slightly, although not markedly, higher than the others, and we have the fullest expression of this change at 37°·5 C. The question now arises what value is to be taken as representing the maximal assimilation at these temperatures? Two different curves (although both of the same type) can be obtained by using either the numbers representing the average of the same number of estimations in each case or the numbers of the first readings only. Either curve gives the true conception of the relation between temperature and assimilation, namely, that when the other conditions are optimal, it is only rise in temperature which renders rise in assimilation possible.

The curve more truly representing the pure effect of the rise in temperature on the assimilation will, however, be that derived from the numbers obtained in the first readings only, for these are the most nearly unaffected by prolonged exposure. At 15° C. and 23°·7 C. the value derived from the first reading is practically the same as the average value; at 30°·5 C. it is ·0157 instead of ·0136; at 37°·5 C., ·0237 instead of ·0163.

The upper curve in fig. 5 is based on these first readings, and must be regarded as the most correct expression of the effect of temperature on assimilation. It much resembles the curve of the effect of temperature upon respiration.

The lower curve, that formed by using the average numbers, is another expression of the same relation, but in this case the depressing influence of comparative high temperatures is also taken into account. It is, therefore, of a more composite nature, and expresses the average effect on a leaf of 6 hours' assimilation at these various temperatures. It is quite evident that by varying the number of assimilation readings entering into the average we could obtain a whole series of curves of the same type.

---

* There is ample evidence that the sudden decrease between the amounts in the first two readings is not in any way due to a choking effect produced by the large amount of carbo-hydrate formed during the first hour, but is a simple temperature effect.

We might even get quite a different temperature to represent the optimal temperature for assimilation, according to the length of time which was allowed to elapse before readings were taken.

Section V.—Summary and Conclusions.

i. *Recapitulation.*

Let us now recall the evidence at our disposal on the question of the relation between temperature and assimilation. In the first place we have the series of curves in figs. 2 and 3 (Section III.). These curves are all of the same type; they show an initial rising portion, changing abruptly into a level line. The greater the intensity of the light, the higher up in the temperature-scale does this change in form occur. The steeply rising portion of the curve is an expression of the fact that, at the temperatures concerned, the intensity of the light is sufficient for maximal assimilation, the long level portion merely gives the amount of CO_2 the light employed was capable of decomposing. From these curves we have obtained the maximal assimilation from $-6°$ C., where the assimilation begins, to $+11°$ C.

We next consider the evidence given by the curve in fig. 4, embodying the results of the experiments in Section IV. (ii.). This is again a curve of the same type as the earlier ones and shows the insufficiency of the eightfold light for maximal assimilation in the middle part of the temperature range. The decided fall at the very high temperatures was a new feature. As each number employed in the curve is the mean of two separate experiments, this steep fall is well established.

It is of interest to compare with this curve the final curve given by Kreusler (1) for the relation between the assimilation and the temperature. The forms are almost identical, thus clearly showing that the curve he obtained was not the true one but was conditioned as to its middle part by insufficiency of the light reaching the leaves.

We now come to the final curve given in fig. 5 (Section IV. (iii.), c.), comprising the range of temperatures from 15–41° C. Here we have full evidence that at every point the assimilation is maximal and the curve obtained rises more steeply the higher the temperature. Above 38° C., however, we have the steep fall, the existence of which was shown in fig. 4.

ii. *Seasonal Variation in the Activity of Leaves.*

On comparing the amounts of assimilation represented in figs. 4 and 5 an apparent anomaly becomes obvious. The actual amounts in fig. 5 are very much smaller than those in fig. 4, although the latter were obviously not maximal at that time. On the other hand, there was conclusive evidence, Section IV. (iii.), c, that in these later experiments the low values represented the maximal assimilation. The explanation is to be found in a seasonal change in the condition of the leaves.

Fig. 4 is based on experiments performed during the winter months, for the main part in February, 1903 ; fig. 5 on experiments carried out in April, 1903. A very short time before the latter experiments, the weather had become decidedly warmer, and a very marked change had taken place in the vegetation generally. This passage of winter into spring is connected with a marked *decrease* in the assimilatory activity of the leaves ; it appears as if at the change of weather they suddenly became sluggish.

The same phenomenon may be noticed on comparing the amounts of assimilation obtained with unit intensity of light in fig. 2 (February, 1902) and fig. 3 (April, 1902). Evidently in April the leaf does not exert its full capability of assimilation, for only ·0020 gramme CO_2 is decomposed under the influence of an intensity of light which in February was sufficient for the breaking up of ·0028 gramme CO_2.

In what way this reduction in the activity is brought about is not yet determined and it is by no means clear what the immediate cause may be. It appears as if in the sluggish condition of the leaves more light were necessary to do the same amount of work.

Frequent experiments with leaves off the same bush must be made at various times of year to throw more light on this seasonal change, and the investigation is in hand. It may be of interest to mention that both in 1901 and 1902 I obtained a low value for the assimilation from October to the middle of December, when the weather was comparatively mild, a high value from the middle of January to March, which was the coldest part of the year, and a low value again in April. The young leaves appear in May, develop slowly during the summer and do not reach a mature state until September.

We have shown that these phenomena do not affect the general form of the curve obtained, but only the absolute amounts representing the maximal assimilation at the various temperatures. The maximal assimilation of a leaf at 30° C. might be ·0240 gramme in February, and in April be ·0136 gramme, but a reduction in the temperature to 11° C. would cause the same proportionate decrease in both cases.

iii. *Synthesis of Complete Curve.*

In synthesising from the figures, given before, the complete curve showing the relation between temperature and assimilation, this effect of the seasonal change on the value of the assimilation must be borne in mind. The experiments represented in figs. 3 and 5 were all carried out in April, during the period of decreased activity. From fig. 3 we obtain the value for the maximal assimilation at all temperatures from — 6° C. to 11° C. ; from fig. 5 for all temperatures from 15° C. to 40°·5 C. These values are placed together in the accompanying diagram (fig. 6), and it will be seen that fig. 5 forms an almost exact continuation of fig. 3. From fig. 4 we know that after 40° C. the assimilation rapidly decreases up to 43° C., we may, therefore,

continue the steep decrease in fig. 6. This gives the complete curve from — 6° C. to +43° C. Therefore, under optimal conditions of light and supply of CO_2, the

Fig. 6.
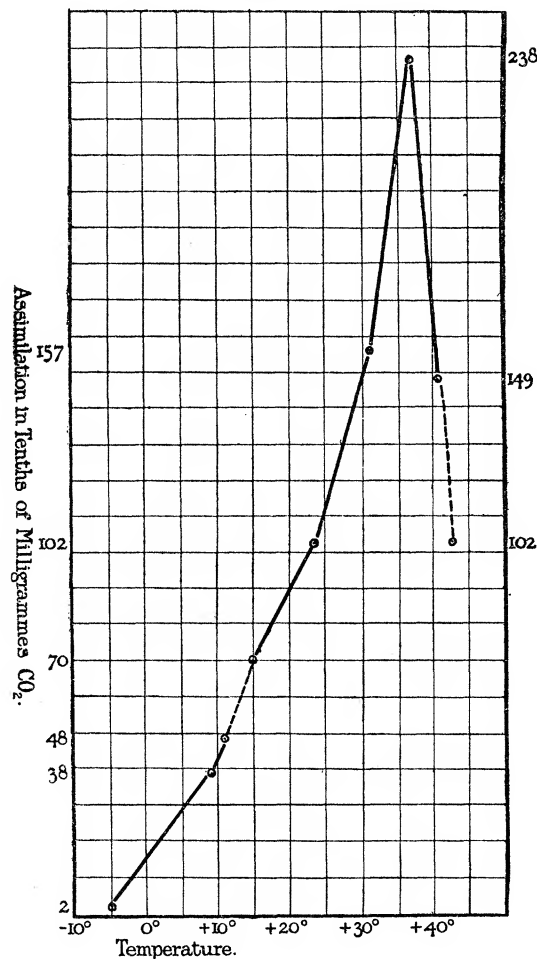
assimilation rises rapidly with rise of temperature until within a few degrees of the temperature fatal to the leaf.

We see then that for every temperature there is a certain intensity of light beyond which further increase in the illumination produces no effect on the assimilation, except indirectly by increasing the internal temperature of the leaf. The influence of the intensity of illumination and of the percentage of CO_2, are therefore of the same secondary nature, and can only affect the assimilation by reducing it if they are below the required optimal amount for the particular temperature.

iv. Conclusions.

(1) *Corresponding to each temperature there is a certain definite amount of assimilation which may be termed the "maximal assimilation for that temperature,"*

this cannot be exceeded, and will not be reached unless both light and CO_2 supply are adequate.

(2) These maximal amounts increase rapidly with increasing temperature, forming a curve which is convex to the temperature abscissa and resembles the curve of the relation between temperature and respiration.

(3) This series of maxima begins to decrease suddenly some degrees below the fatal temperature (*i.e.*, temperature sustainable only for a few hours).

(4) At high temperatures an individual leaf can only maintain a maximal assimilation for a short time. The higher the temperature the sooner the decline begins and the steeper its slope. It follows that a curve of the values of the assimilation at different temperatures in the first hour is not the same as that formed from the values during the second hour, &c.

(5) The assimilatory activity of a mature leaf is not always the same, but varies considerably with the time of year.

BIBLIOGRAPHY.

1. KREUSLER. " Beobachtungen über die Kohlensäure-Aufnahme und Ausgabe der Pflanzen," 'Landwirthschaftliche Jahrbücher,' 1887, p. 711.
2. PFEFFER. 'The Physiology of Plants,' vol. 1, Section 58.
3. BOUSSINGAULT. 'Agronomie,' 1868, vol. 4, p. 312.
4. SAPOSCHNIKOFF. " Über die Grenzen der Anhäufung der Kohlenhydrate," 'Ber. d. D. Bot. Ges.,' 1890, p. 298.
   ——————— " Beitrag zur Kenntniss der Grenzen der Anhäufung von Kohlenhydraten in den Blättern," 'Ber. d. D. Bot. Ges.,' 1893, p. 391.
   ——————— " Eiweisstoffe und Kohlenhydrate der Grünen Blätter als Assimilationsproducte," 'Bot. Centralblatt,' 1895, vol. 63, p. 246.
5. BORODIN. " Untersuchungen über die Pflanzenathmung," 'Mémoires de l'Académie Impériale des Sciences de St. Pétersbourg,' 7 série, vol. 28, p. 4.
6. CLOËZ and GRATIOLET. " Recherches Expérimentales sur la Végétation des Plantes submergées," 'Ann. Sci. Nat.,' S. III., vol. 1, 32, 1851, p. 42.
7. BOUSSINGAULT. 'Agronomie,' vol. 5, p. 16.
8. FRANCOPERT. "Recherches sur la Respiration des Végétaux," 'Comptes Rendus,' 1864, vol. 58, p. 334.
9. BOEHM. 'Sitzungsbericht d. Wien. Akad,' 1873, vol. 1, p. 52.

10. HEINRICH. "Beiträge zur Kenntniss des Temperatur- und Licht-einflusses auf die Sauerstoffausscheidung bei Wasserpflanzen," 'Landwirthschaft. Versuchstationen,' vol. 13, 1871.

11. PFEFFER. " Die Wirkung farbigen Lichtes auf die Zersetzung der Kohlensäure in Pflanzen," 'Arb. d. Bot. Inst. zu Würzburg,' 1874, vol. 1, p. 52.

12. REINKE. " Untersuchungen über die Einwirkung des Lichtes auf die Sauerstoffausscheidung der Pflanzen," 'Bot. Zeit.,' 1884, p. 24.

13. PRJANISCHNIKOW. " Wirkung des Lichtes und der Wärme auf das Ergrünen und auf den Gas-austausch der Pflanzen," Warschau, 1876. 'Botanisches Jahresbericht,' 1876.

14. VAN TIEGHEM. 'Traité de Botanique,' vol. 1, p. 179.

15. JUMELLE. " Recherches physiologiques sur les Lichens," ' Revue Générale de Botanique,' 1892, p. 303.

16. EWART. " Assimilatory Inhibition in Plants," ' Linn. Soc. Journal,' Botany, 1895, vol. 31, p. 389.

17. REINKE. " Untersuchungen über die Einwirkung des Lichtes auf die Sauerstoff-ausscheidung der Pflanzen,' 'Bot. Zeit.,' 1883, p. 713.

18. BOUSSINGAULT. 'Agronomie,' 1868, vol. 4, p. 343.

TABLES OF EXPERIMENTS.

I. *Weight and Area.*—Where two weights are given in this column the first number denotes the weight before the experiment, and the second after. The mean is taken as the true weight of the leaf. When only one is given it is the weight before the experiment.

The assimilation has always been calculated per area, the respiration per weight (see p. 64).

II. *Respiration.*—The values in all the tables, except VII. and XI., are obtained from the data of Table I. (last column) either directly or by interpolation (see the curve of fig. 2).

III. *Apparent Assimilation.*—The apparent assimilation is the amount of CO_2 removed by the leaf in the light from the air-current passing over it, and is measured by the excess of the CO_2 shown by the control tubes over that of the tubes from the chamber. This amount, together with the respiratory CO_2 which the leaf may be considered at that moment as producing, indicates the amount of real assimilation which the leaf is performing.

IV. *Estimations of the* CO_2.—All the amounts are expressed in grammes of CO_2.

V. *Aspirator Rate.*—The rate of flow of the aspirators was always about 800 cub. centims. per hour.

VI. *Percentage of* CO_2.—The percentage of CO_2 employed varied between ·8 and 2·8, and can easily be obtained in any particular case.

$$e.g., \text{per cent. of } CO_2 \text{ by volume} = \frac{\text{weight of } CO_2 \text{ in control tube per hour} \times 500}{8},$$

since the rate of the current is 800 cub. centims. per hour and 500 cub. centims. CO_2 weighs approximately 1 gramme.

VII. *Intensity of Light.*—The light obtained from a single ordinary incandescent mantle when the front of the mantle is 130 millims. from the leaf is termed light of unit intensity, expressed thus— L.In. = 1.

All other intensities of light are measured in terms of this unit—L.In. = 4, &c.

TABLE I.—Respiration Experiments, Temperatures 5°–33° C.

Date.	Temperature.	Number of leaves.	Weight before experiment.	Weight after experiment.	Total amount CO_2 for each 2 hours' reading.					Calculated amount CO_2 per hour per 2 grammes.					Mean value CO_2 per hour per 2 grammes.
					1	2	3	4	5	1	2	3	4	5	
February 4–5, 1902. 2.55 P.M.–2.55 A.M.	5°·8 C.	11	18·30	18·55	·0019	·0019	·0019	·0020½	·0019½	·0001	·0001	·0001	·0001	·0001	·0001
January 31–February 1, 1902 5.20 P.M.–5.20 A.M.	14°·2 C.	8	12·67	12·75	·0031	·0031	·0033	·0032	·0032	·0002½	·0002½	·0002½	·0002½	·0002½	·0002½
February 5–6, 1902. 8.30 P.M.–8.30 A.M.	14°·3 C.	8	12·75	12·80	·0032½	·0032½	·0036	·0036	·0036½	·0002½	·0002½	·0003	·0003	·0003	·0003
January 30–31, 1902 8.20 P.M.–8.30 A.M.	18°·2 C.	7	10·10	10·15	·0052	·0049½	·0045½	—	·0046	·0005	·0005	·0004½	—	·0004½	·0004½
February 6–7, 1902. 8.30 P.M.–8.30 A.M.	18°·1 C.	7	11·77	11·82	·0050½	·0050	·0049	·0048	·0047½	·0004½	·0004	·0004	·0004	·0004	·0004
January 25–26, 1902. 8 A.M.	25°·7 C.	5	7·68	7·72	·0056½	·0052	·0045½	·0045½	·0041	·0007½	·0007	·0006	·0006	·0005½	·0006
January 24, 1902. 11.20 A.M.–11 P.M.	29°·2 C.	4	6·25	6·40	·0055½	·0055½	·0053	·0050½	·0045	·0009	·0009	·0008½	·0008	·0007	·0008½
January 23, 1902. 10.30 A.M.–10.30 P.M.	33°·0 C., went up to 35°·3 C. at end of experiment.	2	3·35	3·37	·0046½	·0047	·0048	·0048	·0051	·0014	·0014	·0014	·0014	·0015	·0014
February 9–10, 1902. 3 P.M.–3 A.M.	33°·1 C.	2	3·05	—	spoilt ·0042	·0042	·0042½	·0041	·0039¾	—	·0014	·0014	·0013¼	·0013½	·0013½

TABLE II.

Assimilation experiments at medium temperatures with unit intensity of light. Single ordinary incandescent burner at low gas pressure (1 inch water). One mantle used throughout.

Weight and area.	Temperature.		Time.	CO_2 supplied. Control tubes.	CO_2 un-assimilated. Tubes from chamber.	Apparent assimilation. Difference.	Respiration for leaf per 2 hours. [Calculated from Table I.]	Real assimilation for leaf for 2 hours.	Real assimilation calculated for 50 sq. cms. and 1 hour.
	Time.	Temp.							
Experiment V.—January 29, 1902. 14°·4 C.									
1st weight, 1·60 grammes.	1.15 P.M.	14·2	1.0 – 2.45 P.M.	Preliminary	Preliminary	—	—	—	—
2nd weight, 1·62 grammes.	3.0 ,,	14·3	2.45– 4.45 ,,	·0331½	·0280½	·0051	·0004	·0055	·0027
Area, 51·5 sq. centims.	4.15 ,,	13·8	4.45– 6.45 ,,	·0336	·0286	·0050	·0004	·0054	·0026½
	7.10 ,,	14·6	6.45– 8.45 ,,	·0331½	·0280	·0051½	·0004	·0055½	·0027
	8.20 ,,	14·5	8.45–10.45 ,,	·0334	·0273	·0061	·0004	·0065	·0031½
	10.45 ,,	14·0	10.45–12.45 A.M.	·0331½	·0275½	·0056	·0004	·0060	·0029
Experiment VI.—January 30, 1902. 18°·0 C.									
1st weight, 1·60 grammes.	11.0 A.M.	18·0	10.15–11.45 A.M.	Preliminary	Preliminary	—	—	—	—
2nd weight, 1·60 grammes.	12.45 P.M.	18·0	11.45– 1.45 P.M.	·0323	·0281	·0042	·0007	·0049	·0027
Area, 44·8 sq. centims.	3.40 ,,	17·8	1.45– 3.45 ,,	·0323½	·0280½	·0043	·0007	·0050	·0027½
	4.40 ,,	18·1	3.45– 5.45 ,,	·0322½	·0280	·0042½	·0007	·0049½	·0027
	6.45 ,,	18·2	5·45– 7.45 ,,	·0330½	·0286½	·0044	·0007	·0051	·0028
	8.20 ,,	18·2							
Experiment VII.—January 25, 1902. 25°·7 C.									
1st weight, 1·63 grammes.	11.0 A.M.	25·6	10.15–11.20 A.M.	Preliminary	Preliminary	—	—	—	—
2nd weight, 1·64 grammes.	12.0 noon	25·6	11.20– 1.20 P.M.	·0327	·0278½	·0048½	·0011	·0059½	·0029½
Area, 50·0 sq. centims.	1.0 P.M.	25·6	1.20– 3.20 ,,	·0323½	·0273	·0050½	·0011	·0061½	·0030½
	4.0 ,,	25·6	3.20– 5.20 ,,	·0321	·0268	·0053	·0011	·0064	·0032
	7.30 ,,	25·8	5.20– 7.20 ,,	·0321	·0270	·0051	·0011	·0062	·0031
Experiment VIII.—January 23, 1902. 29°·1 C.									
1st weight, 1·38 grammes.	10.40 A.M.	29·0	10.30–12.20 P.M.	Preliminary	Preliminary	—	—	—	—
2nd weight, 1·36 grammes.	12.45 P.M.	29·1	12.20– 2.20 ,,	·0333½	·0297½	·0036	·0012	·0048	·0028
Area, 43·0 sq. centims.	2.30 ,,	29·2	2.20– 4·20 ,,	·0329	·0294	·0035	·0012	·0047	·0027½
	4.50 ,,	29·2	4.20– 6.20 ,,	·0333	·0292½	·0040½	·0012	·0052½	·0030½
	8.0 ,,	29·2	6.20– 8·20 ,,	·0323½	·0288½	·0035	·0012	·0047	·0027½
Experiment IX.—February 1, 1902. 33°·1 C.									
1st weight, 1·45 grammes.	12.40 P.M.	33·0	10.45–12.45 P.M.	Preliminary	Preliminary	—	—	—	—
Area, 43·5 sq. centims.	1.0 ,,	33·0	12.45– 2.45 ,,	·0317½	·0288	·0029½	·0020	·0049½	·0028½
	3·20 ,,	33·2	2.45– 4.45 ,,	·0324½	·0295	·0029½	·0020	·0049½	·0028½

Subsequent readings spoilt.

TABLE III.

Further assimilation experiments at medium temperatures with unit intensity of light. Single incandescent burner at low gas pressure (1 inch water). One mantle employed throughout.

Weight and area.	Temperature. Time.	Temperature. Temp.	Time.	CO_2 supplied. Control tubes.	CO_2 unassimilated. Tubes from chamber.	Apparent assimilation. Difference.	Respiration for leaf per 2 hours. [Calculated from Table I.]	Real assimilation for leaf for 2 hours.	Real assimilation calculated for 50 sq. cms., and 1 hour.
Experiment X.—February 8, 1902. 5°·5 C.									
Weight, 1·60 grammes. Area, 41·5 sq. centims.	10.45 A.M.	5·5	10.45–11.50 A.M.	Preliminary	Preliminary	—	—	—	—
	12 noon	5·5	11.15– 1.15 P.M.	·0304½	·0258	·0046½	·0001½	·0048	·0029
	2.0 P.M.	5·6	1.15– 3.15 ,,	·0316½	·0270	·0046½	·0001½	·0048	·0029
			3.15– 5.15 ,,	·0313	·0269	·0044	·0001½	·0045½	·0027½
Experiment XI.—February 6, 1902. 10°·2 C.									
Weight, 1·70 grammes. Area, 49·8 sq. centims.	10.40 A.M.	10·0	10.20–11.50 A.M.	Preliminary	Preliminary	—	—	—	—
	2.10 P.M.	10·1	11.50– 1.50 P.M.	·0254	·0208½	·0045½	·0002½	·0048	·0024*
	4.0 ,,	10·2	1.50– 3.50 ,,	·0297½	·0241	·0056½	·0002½	·0059	·0028½
	7.40 ,,	10·4	3.50– 5.50 ,,	·0325	·0271½	·0053½	·0002½	·0056	·0028
Experiment XII.—February 7, 1902. 10°·1 C.									
Weight, 1·60 grammes. Area, 42·5 sq. centims.	10.0 A.M.	10·05	9.50– 2.30 P.M.	Preliminary	Preliminary	—	—	—	—
	4.30 P.M.	10·05	2.30– 4.30 ,,	·0325	·0279½	·0045½	·0002½	·0048	·0028
	5.5 ,,	10·05	4.30– 6.30 ,,	·0327	·0281½	·0045½	·0002½	·0048	·0028
	8.40 ,,	10·1	6.30– 8.30 ,,	·0334	·0288	·0046	·0002½	·0048½	·0028½
Experiment XIII.—February 5, 1902. 14°·2 C.									
1st weight, 1·45 grammes. 2nd weight, 1·42 grammes. Area, 43·2 sq. centims.	10.30 A.M.	14·2	10.10–11.50 A.M.	Preliminary	Preliminary	—	—	—	—
	12 noon	14·2	11.50– 1.50 P.M.	·0339½	·0283½	·0056	·0004	·0060	·0035
	2.45 P.M.	14·2	1.50– 3.50 ,,	·0331½	·0276½	·0055	·0004	·0059	·0034½
	5.45 ,,	14·2	3.50– 5.50 ,,	·0337	·0280½	·0056½	·0004	·0060½	·0035
Experiment XIV.—February 11, 1902. 14°·0 C.									
Weight, 1·60 grammes. Area, 45·3 sq. centims.	12 noon	14·05	12.0 – 2.0 P.M.	Preliminary	Preliminary	—	—	—	—
	2.45 P.M.	14·05	2.0 – 4.0 ,,	·0318½	·0274	·0044½	·0004	·0048½	·0027
	3.45 ,,	14·05	4.0 – 6.0 ,,	—	—	—	—	—	—
	6.35 ,,	13·9	6.0 – 8.0 ,,	·0318	·0276½	·0041½	·0004	·0045½	·0025
Experiment XV.—February 10, 1902. 25°·0 C.									
Weight, 1·55 grammes. Area, 45·7 sq. centims.	9.30 A.M.	25·0	9.30–11.30 A.M.	Preliminary	Preliminary	—	—	—	—
	12.30 P.M.	24·9	11.30– 1.30 P.M.	·0316½	·0266½	·0050	·0010½	·0060½	·0032
	3.15 ,,	25·0	1.30– 3.30 ,,	·0321	·0272½	·0048½	·0010½	·0059	·0031
	6.15 ,,	25·4	3.30– 5.30 ,,	·0324	·0271	·0053	·0010½	·0063½	·0035

* At end of reading, a streak was noticed on window of bath, and to this the smallness of the amount is due.

TABLE IV.

Assimilation experiments at low temperatures with unit intensity of light.   Single incandescent burner at low gas pressure (1 inch water).   A new mantle for each experiment.

Weight and area.	Temperature.	Time.	CO₂ supplied. Control tubes.	CO₂ un-assimilated. Tubes from chamber.	Apparent assimilation. Difference.	Respiration for leaf per 2 hours. [Calculated from Table I.]	Real assimilation for leaf for 2 hours.	Real assimilation calculated for 50 sq. cms., and 1 hour.
		Experiment XVI.—February 27, 1902. +1° C.						
1st weight, 1·68 grammes. 2nd weight, 1·63 grammes. Area, 43·8 sq. centims.	+ 0·9 to 1·1° C.	10.35–12.30 P.M. 12.30– 2.30 „ 2.30– 4.30 „ 4.30– 6.30 „	Preliminary ·0310 ·0293 ·0305½	Preliminary ·0274 ·0256½ ·0272½	— ·0036 ·0036½ ·0033	— ·0000½ ·0000½ ·0000½	— ·0036½ ·0037 ·0033½	— ·0020½ ·0021 ·0019
		Experiment XVII.—March 2, 1902. +2° C.						
1st weight, 1·80 grammes. 2nd weight, 1·81 grammes. Area, 46·0 sq. centims.	+ 2° C. throughout.	10. 5–11.35 A.M. 11.35– 1.35 P.M. 1.35– 3.35 „ 3.35– 5.35 „	Preliminary ·0270½ ·0258 ·0269½	Preliminary ·0228 ·0213½ ·0225	— ·0042½ ·0044½ ·0044½	— ·0000½ ·0000½ ·0000½	— ·0043 ·0045 ·0045	— ·0023½ ·0024½ ·0024½
		Experiment XVIII.—February 13, 1902. +2°·4.						
1st weight, 1·82 grammes. 2nd weight, 1·82 grammes. Area, 49·0 sq. centims.	Generally, 2·4° C. Lowest temperature noted, 2° C. Highest, 2·5° C.	11.15–12.45 P.M. 12.45– 2.45 „ 2.45– 4.45 „ 4.45– 6.45 „	Preliminary ·0336½ ·0330 ·0331	Preliminary ·0285½ ·0278½ ·0281½	— ·0051 ·0051½ ·0049½	— ·0000½ ·0000½ ·0000½	— ·0051½ ·0052 ·0050	— ·0026 ·0026½ ·0025½
		Experiment XIX.—March 7, 1902. 3°·6 C.						
1st weight, 1·40 grammes. 2nd weight, 1·45 grammes. Area, 45·8 sq. centims.	+ 3°·6 C. throughout.	10.20–11.50 A.M. 11.50– 1.50 P.M. 1.50– 3.50 „ 3.50– 5.50 ·,	Preliminary ·0301 ·0297½ ·0296½	Preliminary ·0250 ·0246 ·0245	— ·0051 ·0051½ ·0051½	— ·0000½ ·0000½ ·0000½	— ·0051½ ·0052 ·0052	— ·0028½ ·0028½ ·0028½
	Temperature allowed to rise. 7.12 P.M., 8°·7 C. 10. 0 „ 9°·3 C. 1. 0 A.M., 9°·2 C.	5.50– 7.50 P.M. 7.50– 9.50 „ 9.50–11.50 „	·0302 ·0294½ ·0284½	·0253 ·0242 ·0235	·0049 ·0052½ ·0049½	·0002½ ·0002½ ·0002½	·0051½ ·0055 ·0052	·0028½ ·0030 ·0028½
		Experiment XX.—March 4, 1902. 8°·8 C.						
1st weight, 1·68 grammes. 2nd weight, 1·75 grammes. Area, 48·2 sq. centims.	10.30 A.M., 8°·9 C. 11.15 „ 8°·6 C. 2.30 P.M., 8°·9 C.	10.15–11.45 A.M. 11.45– 1.45 P.M. 1.45– 3.45 „	Preliminary ·0232 ·0218	Preliminary ·0176 ·0163	— ·0056 ·0055	— ·0002½ ·0002½	— ·0058½ ·0057½	— ·0030 ·0029½

TABLE IV.—*continued.*

Weight and area.	Temperature.	Time.	CO₂ supplied. Control tubes.	CO₂ un-assimilated. Tubes from chamber.	Apparent assimilation. Difference.	Respiration* for leaf per 2 hours. [Calculated from Table I.]	Real assimilation for leaf for 2 hours.	Real assimilation calculated for 50 sq. cms., and 1 hour.
			Experiment XXI.—March 1, 1902. − 6°·0 C.					
1st weight, 1·85 grammes. Area, 49·0 sq. centims.	About − 6°·0 C. the whole time.	10.30– 1.0 P.M. 1.0 – 2.15 ,, 2.15– 4.15 ,, 4.15– 6.15 ,,	} Preliminary ·0303½ ·0311½	Preliminary ·0299 ·0304½	— ·0004½ ·0007	— — —	— ·0004½ ·0007	— ·0002½ ·0003½
			Experiment XXII.—February 24, 1902. − 3°·6 C.					
1st weight, 1·78 grammes. Area, 48·7 sq. centims.	− 3°·6 C. to −3°·8 C., with a few minutes at − 0·5 C. in first reading.	10.10–11.45 A.M. 11.45– 1.45 P.M. 1.45– 3.45 ,, 3.45– 5.45 ,,	Preliminary ·0303½ ·0300½ ·0312½	Preliminary ·0282 ·0283 ·0297	— ·0021½ ·0017½ ·0015½	— — —	— ·0021½ ·0017½ ·0015½	— ·0011 ·0009 ·0003
			Experiment XXIII.—February 20, 1902. − 1°·0 C.					
Weight, 1·66 grammes. Area, 44·0 sq. centims.	− 2°·0 C. − 1°·0 C. − 1°·0 C.	10.45–12.15 P.M. 12.15– 2.15 ,, 2.15– 4.15 ,, 4.15– 6.15 ,,	Preliminary ·0300½ ·0307½ ·0314½	Preliminary ·0278½ ·0280 ·0288	— ·0022 ·0027½ ·0026½	— — —	— ·0022 ·0027½ ·0026½	— ·0012½ ·0015½ ·0015
			Experiment XXIV.—February 17, 1902. − 1°·5 C.					
1st weight, 1·67 grammes. 2nd weight, 1·67 grammes. Area, 43·7 sq. centims.	− 1°·4 C. to 1°·6 C.	12.0 – 2.0 P.M 2.0 – 4.0 ,, 4.0 – 6.0 ,, 6.0 – 8.0 ,,	Preliminary ·0317 ·0334 ·0338	Preliminary ·0299½ ·0314 ·0320	— ·0017½ ·0020 ·0018	— — —	— ·0017½ ·0020 ·0018	— ·0010 ·0011½ ·0010

* The respiration at temperatures below 0° C. amounts to less than the error of observation, and has therefore been neglected.

Table V.

Assimilation experiments at low and medium temperatures, with various low intensities of light. Single incandescent burner at low gas pressure (1 inch water). New mantle each experiment.

Intensity of light.	Weight and area.	Temperature.	Time.	CO_2 supplied. Control tubes.	CO_2 un-assimilated. Tubes from chamber.	Apparent assimilation. Difference.	Apparent assimilation calculated for 50 sq. cms., and 1 hour.	Real assimilation* calculated for 50 sq. cms., and 1 hour.
			Experiment XXVIII.—May 3, 1902. 0°·4 C.					
Unit.	1st weight, 1·68 grammes. 2nd weight, 1·70 grammes. Area, 40·4 sq. centims.	0°·4 C. to 0°·3 C. throughout.	9.30–11.0 A.M. 11. 0– 1.0 P.M. 1. 0– 3.0 ,,	Preliminary ·0295½ ·0305	Preliminary ·0269½ ·0274	— ·0026 ·0031	— ·0016 ·0019	— ·0016 ·0019
Twofold at 3 P.M.			3.10–4.0 P.M. 4. 0–6.0 ,, 6. 0–8.0 ,,	Preliminary ·0308 ·0300	Preliminary ·0272 ·0270	— ·0036 ·0030	— ·0022 ·0018½	— ·0022 ·0018½
			Experiment XXIX.—April 12–13, 1902. 8°·8 C.					
Unit.	1st weight, 1·80 grammes. Area, 44·6 sq. centims.	8°·8 C. throughout	12.30–2.0 P.M. 2. 0–4.0 ,, 4. 0–6.0 ,, 6. 0–8.0 ,,	Preliminary ·0313 ·0320 Spoilt	Preliminary ·0275½ ·0283 —	— ·0037½ ·0037	— ·0021 ·0020½	— ·0023 ·0022½
Twofold at 8 P.M.	2nd weight, 1·80 grammes.	8°·8 C. at 8 P.M. 9° C. at 9 A.M.	8. 0– 9.40 P.M. 9.40–11.40 ,, 11.40– 1.40 A.M. 1.40– 3.40 ,, 3.40– 5.40 ,, 5.40– 7.40 ,,	Preliminary ·0331 ·0332½ ·0328 ·0331 ·0328	Preliminary ·0264½ ·0266½ ·0263½ ·0266 ·0263	— ·0066½ ·0066 ·0064½ ·0065 ·0065	— ·0037 ·0037 ·0036 ·0036½ ·0036½	— ·0039 ·0039 ·0038 ·0038½ ·0038½
			Experiment XXX.—April 13–14, 1902. 9°·2 C.					
Unit.	1st weight, 1·58 grammes. Area, 46·1 sq. centims.	9°·1 C. at 7 P.M.	6.50– 9.15 P.M. 9.15–11.15 ,, 11.15– 1.15 A.M. 1.15– 3.15 ,, 3.15– 5.15 ,, 5.15– 7.15 ,, 7.15– 9.40 ,,	Preliminary ·0281½ ·0284½ ·0280 — — —	Preliminary ·0248½ ·0250½ ·0246 — — —	— ·0033 ·0034 ·0034 — — —	— ·0018 ·0018½ ·0018½ — — —	— ·0020 ·0020½ ·0020½ — — —
Fourfold at 9.40 A.M.		9°·2 C. at 9 A.M. 9°·4 C. at 7 P.M.	9.40–10.50 A.M. 11.50–12.50 P.M. 12.50– 2.50 ,, 2.50– 4.50 ,, 4.50– 6.50 ,,	Preliminary ·0444 ·0444½ ·0441½ ·0440½	Preliminary ·0379½ ·0380 ·0380 ·0374	— ·0064½ ·0064½ ·0061⅓ ·0066½	— ·0035 ·0035 ·0033 ·0036	— ·0037 ·0037 ·0035 ·0038

* Respiration calculated from Table I.

TABLE V.—continued.

Intensity of light.	Weight and area.	Temperature. Time.	Temperature. Temp.	Time.	CO₂ supplied. Control tubes.	CO₂ un-assimilated. Tubes from chamber.	Apparent assimila-tion. Difference.	Apparent assimila-tion calculated for 50 sq. cms., and 1 hour.	Real assimila-tion* calculated for 50 sq. cms., and 1 hour.
				Experiment XXXI.—April 25, 1902. 11°·4 C.					
Unit.	Area, 45·1 sq. centims. 1st weight, 1·65 grammes.	9.15 P.M. 1 A.M.	11·4 11·4	9.10–10.30 P.M. 10.30–12.30 A.M. 12.30– 2.30 ,, 2.30– 4.30 ,, 4.30– 6.30 ,, 6.30– 8.30 ,,	Preliminary ·0306½ ·0312 ·0309½ ·0311½ ·0306½	Preliminary ·0275½ ·0275 ·0275½ ·0270 ·0268½	— ·0031 ·0037 ·0034 ·0041½ ·0038	— ·0017 ·0020½ ·0019 ·0023 ·0021	— ·0019 ·0022½ ·0021 ·0025 ·0023
Sixfold at 9.15 A.M.		9.15 A.M. 1 P.M.	11·4 11·4	9.15–10.25 A.M. 10.25–12.25 P.M. 12.25– 2.25 ,,	Preliminary ·0325 ·0318	Preliminary ·0237½ ·0230	— ·0087½ ·0088	— ·0048½ ·0049	— ·0050½ ·0051
Fourfold at 2.30 P.M.	2nd weight, 1·70 grammes.	6 P.M. 8 ,,	11·4 11·4	2.30–3.15 P.M. 3.15–5.15 ,, 5.15–7.15 ,,	Preliminary ·0316½ ·0312	Preliminary ·0235½ ·0231	— ·0081 ·0081	— ·0045 ·0045	— ·0046½ ·0046½
				Experiment XXXII.—April 26, 1902. 11°·4 C.					
Unit.	Area, 36·6 sq. centims. 1st weight, 1·50 grammes.	9 P.M. Midnight	11·4 11·4	9.0 –10.40 P.M. 10.40–12.40 A.M. 12.40– 2.40 ,, 2.40– 4.40 ,, 4.40– 6.40 ,, 6.40– 8.40 ,,	Preliminary ·0308½ ·0308 ·0307 ·0306 —	Preliminary ·0281½ ·0279½ ·0277 ·0275 —	— ·0027 ·0028½ ·0030 ·0031	— ·0018½ ·0019½ ·0020½ ·0021	— ·0020½ ·0021½ ·0022½ ·0023
Twofold at 8.40 A.M.	2nd weight, 1·50 grammes.	8.40 A.M. 11. 0 ,, 5.40 P.M.	11·4 11·4 11·4	8.40– 9.40 A.M. 9.40–11.40 ,, 11.40– 1.40 P.M. 1.40– 3.40 ,, 3.40– 5.40 ,,	Preliminary ·0304½ ·0314 ·0315 ·0316	Preliminary ·0254 ·0261 ·0266 ·0262½	— ·0050½ ·0053 ·0049 ·0053½	— ·0035 ·0036 ·0033½ ·0036½	— ·0037 ·0038 ·0035½ ·0038½
				Experiment XXXIII.—April 24 and 25, 1902. 11°·4 C.					
Intensity = 1·37.	Area, 46·5 sq. centims. 1st weight, 1·65 grammes.	6.45 P.M. 10.0 ,, 1.0 A.M. 8.15 ,,	11·4 11·4 11·4 11·5	6.45– 8.45 P.M. } 8.45–10.45 ,, } 10.45–12.45 A.M. 12.45– 2.45 ,, 2.45– 4.45 ,, 4.45– 6.45 ,, 6.45– 8.45 ,,	Preliminary ·0309½ ·0310 ·0308½ — —	Preliminary ·0259 ·0259 ·0258½ — —	— ·0050½ ·0051 ·0050 — —	— ·0027 ·0027½ ·0027 — —	— ·0029 ·0029½ ·0029 — —
Fourfold at 10.15 A.M.	2nd weight, 1·72 grammes.	10.20 A.M. 12.0 noon 3.30 P.M.	11·3 11·3	8.45–11.25 A.M. 11.25– 1.25 P.M. 1.25– 3.25 ,,	Preliminary ·0335 ·0340½	Preliminary ·0252½ ·0257½	— ·0082½ ·0083	— ·0045 ·0045	— ·0046½ ·0046½

* Respiration calculated from Table I.

TABLE V.—*continued.*

Intensity of light.	Weight and area.	Temperature. Time.	Temp.	Time.	CO_2 supplied. Control tubes.	CO_2 un-assimilated. Tubes from chamber.	Apparent assimilation. Difference.	Apparent assimilation calculated for 50 sq. cms., and 1 hour.	Real assimilation* calculated for 50 sq. cms., and 1 hour.
				Experiment XXXIV.—April 29, 1902. 25°·2 C.					
Unit.	Area, 41·5 sq. centims. 1st weight, 1·60 grammes.	10.15 A.M. 12.0 noon 1.15 P.M.	25·2 25·2 25·2	10.15–11.15 A.M. 11.15–12.15 P.M. 12.15– 1.15 ,,	Preliminary ·0159½ ·0152	Preliminary ·0146½ ·0139	— ·0013 ·0013	— ·0015½ ·0015½	— ·0021½ ·0021½
Fourfold at 1.15 P.M.		1.45 P.M. 3. 0 ,, 4. 0 ,,	25·2 25·2 25·2	1.15–2.0 P.M. 2. 0–3.0 ,, 3. 0–4.0 ,,	Preliminary ·0146½ ·0152	Preliminary ·0098½ ·0108	— ·0048 ·0044	— ·0058 ·0053	— ·0064 ·0059
				Experiment XXXV.—April 30, 1902. 25°·2 C.					
Unit.	Area, 40·0 sq. centims. 1st weight, 1·75 grammes.			12.0–3.0 P.M.		No	readings	taken.	
Twofold at 3 P.M.	2nd weight, 1·78 grammes.	3.0 P.M. 5.0 ,,	25°·2 25 ·2	3.0–4.0 P.M. 4.0–5.0 ,, 5.0–6.0 ,,	Preliminary ·0125 ·0122	Preliminary ·0098½ ·0098	— ·0026½ ·0024	— ·0033 ·0030½	— ·0040 ·0036½
				Experiment XXXVI.—April 19 and 20, 1902. 24°·8 C.					
Fourfold.	Area, 39·2 sq. centims. 1st weight, 1·54 grammes.	1. 0 P.M. 2. 0 ,, 6.45 ,,	24·8 24·8 24·8	12.0– 2.0 P.M. 2.0– 4.0 ,, 4.0– 6.0 ,, 6.0– 8.0 ,, 8.0–10.0 ,,	Preliminary ·0302 ·0301½ ·0301 ·0298	Preliminary ·0235½ ·0233½ ·0236½ ·0231½	— ·0066½ ·0068 ·0064½ ·0066½	— ·0042½ ·0043½ ·0041 ·0042½	— ·0048 ·0049½ ·0047½ ·0048
Unit at 10 P.M.	2nd weight, 1·60 grammes.	10.15 P.M. 10.0 A.M.	24·8 25·0	10.0–12.0 night 12.0– 2.0 A.M. 2.0– 4.0 ,, 4.0– 6.0 ,, 6.0– 8.0 ,, 8.0–10.0 ,,	Preliminary ·0295 ·0299½ ·0305 ·0301 ·0298	Preliminary ·0278½ ·0282½ ·0290 ·0281½ ·0279	— ·0016½ ·0017 ·0015 ·0019½ ·0019	— ·0010½ ·0011 ·0009½ ·0012½ ·0012	— ·0017 ·0017½ ·0016 ·0020 ·0019½

* Respiration calculated from Table I

TABLE VI.—Summary of the Experiments in Table V., setting forth the Ratios of the Assimilation with the different Intensities of Light. [The Absolute Amounts of Assimilation given are the Means of the Readings in the Respective Experiments of Table V.]

Number of experiment.	Temp.	Apparent assimilation 50 sq. centims. 1 hour.				Real assimilation 50 sq. centims. 1 hour.				Ratios of real assimilations.			
		L.In. = 1.	L.In. = 2.	L.In. = 4.	L.In. = 6.	L.In. = 1.	L.In. = 2.	L.In. = 4.	L.In. = 6.	L.In. = 1.	L.In. = 2.	L.In. = 4.	L.In. = 6.
XXVIII.	0°·4 C.	·0017½	·0020	—	—	·0017½	·0020	—	—	1	1·14	—	—
XXIX.	8°·8 C.	·0020½	·0036½	—	—	·0022½	·0038½	—	—	1	1·71	—	—
XXX.	9°·2 C.	·0018½	—	·0035	—	·0020½	—	·0036½	—	1	—	1·78	—
XXXI.	11°·4 C.	·0020	—	·0045	·0049	·0022	—	·0046½	·0050½	1	—	2·16	2·34
XXXII.	11°·4 C.	·0020	·0035	—	—	·0022	·0037½	—	—	1	1·70	—	—
XXXIII.	11°·4 C.	—	—	·0045	—	—	—	·0046½	—	1	—	—	—
XXXIV.	25°·2 C.	·0015½	—	·0055½	—	·0021½	—	·0061½	·0063	1	—	2·88	—
XXXV.	25°·2 C.	—	·0032	—	—	—	·0038½	—	—	1	1·80	—	—
XXXVI.	24°·8 C.	·0013	—	·0042½	—	·0018½	—	·0048½	—	1	—	2·80	—

TABLE VII.

Assimilation experiments at medium and high temperatures with light of eightfold intensity (L.In. = 8). Two ordinary incandescent burners at low gas pressure. New mantles for each experiment.

Illumina-tion.	Weight and area.	Temperature.		Time.	CO_2 supplied.	CO_2 in current after leaving chamber.	Apparent assimila-tion per 2 hours.	Respira-tion per 2 hours.	Real assimila-tion per hour per 50 sq. cms.
		Time.	Temp.						
Experiment XXXVII.—March 4, 1903. 11°·0 C.									
Light.	Area, 38·8 sq. centims. Weight, 1·50 grammes.	11° C. throughout.		10.0 –11.30 A.M. 11.30– 1.30 P.M. 1.30– 3.30 ,, 3.30– 5.30 ,,	Preliminary ·0366 ·0378 ·0368	Preliminary ·0258 ·0270 ·0260	— ·0108 ·0108 ·0108		— — —
				Mean values per hour			·0054	·0002*	·0072
Intensity changed to L.In.=4·6.				5.30– 7.30 P.M. 7.30– 9.30 ,, 9.30–11.30 ,,	Preliminary ·0377 ·0380	Preliminary ·0275 ·0283	— — ·0102 ·0097		— — —
				Mean values per hour			·0050	·0002*	·0066
Experiment XXXVIII.—January 29 and 30, 1903. 25°·4 C.									
Darkened.	Area, 41·0 sq. centims. 1st weight, 1·70 grammes.	10 A.M.	25°·3	10.0 –11.30 A.M. 11.30– 1.30 P.M. 1.30– 3.30 ,,	Preliminary ·0225 ·0220	Preliminary ·0233½ ·0226		— ·0008½ ·0006	
Light 4 P.M.		4 P.M. 11.30 P.M. midnight.	25·3 26·4 25·7	4.0 – 6.0 P.M. 6.0 – 9.0 ,, 9.0 –12.0 night 12.0 – 3.0 A.M. 3.0 – 6.0 ,, 6.0 – 9.0 ,,	Preliminary ·0416 ·0427 ·0410 ·0419 ·0403	Preliminary ·0128 ·0160 ·0168 ·0184 ·0199	— ·0288 ·0267 ·0242 ·0235 ·0204	— — — — — —	— — — — — —
Darkened.	2nd weight, 1·75 grammes.	9 A.M.	25·4	9.0 –11.0 A.M. 11.0 – 1.0 P.M. 1.0 – 3.0 ,, 3.0 – 5.0 ,,	Preliminary ·0230 ·0228 ·0222	Preliminary ·0272 ·0266 ·0258		— ·0042 ·0038 ·0036	
				Mean values per hour			·0123½	·0011½	·0128

* Calculated from Table I.

TABLE VII.—*continued*.

Illumination.	Weight and area.	Temperature. Time.	Temp.	Time.	CO₂ supplied.	CO₂ in current after leaving chamber.	Apparent assimilation per 2 hours.	Respiration per 2 hours.	Real assimilation per hour per 50 sq. cms.

Experiment XXXIX.—January 31, 1903.   25°·3 C.

Illumination.	Weight and area.	Time.	Temp.	Time.	CO₂ supplied.	CO₂ in current after leaving chamber.	Apparent assimilation per 2 hours.	Respiration per 2 hours.	Real assimilation per hour per 50 sq. cms.
Darkened.	Area, 37·2 sq. centims. 1st weight, 1·50 grammes.	10 A.M.	25°·1	10–11.30 A.M. 11.30–1.30 P.M. 1.30–3.30 ,,	Preliminary ·0333 ·0340	Preliminary ·0352 ·0346		— ·0019 ·0006	
Light 4 P.M.		4 P.M. 5 P.M.	25·1 25·5	4–6 P.M. 6–8 ,, 8–10 ,, 10–12 night	Preliminary ·0361 ·0373 ·0372	Preliminary ·0186 ·0218 ·0215	— ·0175 ·0155 ·0157	— — —	— — —
Darkened midnight.	2nd weight, 1·55 grammes.	9 A.M.	25·5	12–2 A.M. 2–4 ,, 4–6 ,,	Preliminary ·0362 ·0356	Preliminary ·0392 ·0392		— ·0030 ·0036	
					Mean values per hour		·0081	·0011½	·0125

Experiment XL.—February 4, 1903.   32°·1 C.

Illumination.	Weight and area.	Time.	Temp.	Time.	CO₂ supplied.	CO₂ in current after leaving chamber.	Apparent assimilation per 2 hours.	Respiration per 2 hours.	Real assimilation per hour per 50 sq. cms.
Darkened.	Area 40·0 sq. centims. 1st weight, 1·52 grammes.	10.30 A.M. 12 noon	32°·1 32·1	10.30–12 noon 12–2 P.M. 2–4 ,,	Preliminary ·0337 ·0336	Preliminary ·0372 ·0362		— ·0035 ·0026	
Light 4 P.M.		4 P.M. 6 P.M.	32·1 32·5	4–6 P.M. 6–8 ,, 8–10 ,, 10–12 night	Preliminary ·0353 ·0363 ·0356	Preliminary ·0173 ·0183 ·0199	— ·0180 ·0180 ·0157	— — —	— — —
Darkened midnight.	2nd weight, 1·52 grammes.	9 A.M.	32·1	12–2 A.M. 2–4 ,, 4–6 ,,	Preliminary ·0354 ·0361	Preliminary ·0396 ·0396		— ·0042 ·0035	
					Mean values per hour		·0086	·0018½	·0129½

Experiment XLI.—February 5, 1903.   32°·2 C.

Illumination.	Weight and area.	Time.	Temp.	Time.	CO₂ supplied.	CO₂ in current after leaving chamber.	Apparent assimilation per 2 hours.	Respiration per 2 hours.	Real assimilation per hour per 50 sq. cms.
Darkened.	Area 35·0 sq. centims. 1st weight, 1·40 grammes.	10 A.M. 1 P.M.	32°·1 32·1	10–11.30 A.M. 11.30–1.30 P.M. 1.30–3.30 ,,	Preliminary ·0334 ·0336	Preliminary ·0363 ·0351		— ·0029 ·0015	
Light 3.30 P.M.		4 P.M. 5.30 P.M.	32·3 32·3	3.30–5.30 P.M. 5.30–7.30 ,, 7.30–9.30 ,, 9.30–11.30 ,,	Preliminary ·0321 ·0320 ·0315	Preliminary ·0169 ·0175 ·0167	— ·0152 ·0145 ·0148	— — —	— — —
Darkened 11.30 P.M.	2nd weight, 1·40 grammes.	9.30 A.M.	32·3	11.30–1.30 A.M. 1.30–3.30 ,, 3.30–5.30 ,,	Preliminary ·0325 ·0315	Preliminary ·0370 ·0362		— ·0015 ·0047	
					Mean values per hour		·0074	·0017	·0130

TALLE VII.—*continued.*

Illumina-tion.	Weight and area.	Temperature.		Time.	CO₂ supplied.	CO₂ in current after leaving chamber.	Apparent assimila-tion per 2 hours.	Respira-tion per 2 hours.	Real assimila-tion per hour per 50 sq. cms.
		Time.	Temp.						

Experiment XLII.—February 2, 1903.   38° 3 C.

Illumina-tion.	Weight and area.	Time.	Temp.	Time.	CO₂ supplied.	CO₂ in current after leaving chamber.	Apparent assimila-tion per 2 hours.	Respira-tion per 2 hours.	Real assimila-tion per hour per 50 sq. cms.
Darkened.	Area, 38·7 sq. centims. 1st weight, 1·40 grammes.	10 A.M. 12 noon	38·1 38·1	10.0 –11.30 A.M. 11.30– 1.30 P.M. 1.30– 3.30 ,,	Preliminary ·0359 ·0368	Preliminary ·0402 ·0400		— ·0043 ·0032	
Light 3.30 P.M.		3.30 P.M. 5.30 ,,	38·4 38·4	3.30– 5.30 P.M. 5.30– 7.30 ,, 7.30– 9.30 ,, 9.30–11.30 ,,	Preliminary ·0378 ·0379 ·0367	Preliminary ·0242 ·0231 ·0240	— ·0136 ·0148 ·0127	— — — —	— — — —
Darkened 11.30 P.M.	2nd weight, 1·40 grammes.	9 A.M.	38·3	11.30– 1.30 A.M. 1.30– 3.30 ,, 3.30– 5.30 ,,	Preliminary ·0356 ·0347	Preliminary ·0408 ·0385		— ·0052 ·0038	
				Mean values per hour			·0068½	·0020½	·0115

Experiment XLIII.—February 3, 1903.   38° 3 C.

Illumina-tion.	Weight and area.	Time.	Temp.	Time.	CO₂ supplied.	CO₂ in current after leaving chamber.	Apparent assimila-tion per 2 hours.	Respira-tion per 2 hours.	Real assimila-tion per hour per 50 sq. cms.
Darkened.	Area, 39·7 sq. centims. 1st weight, 1·48 grammes.	10 A.M. 12 noon	38·3 38·3	10.0 –11.30 A.M. 11.30– 1.30 P.M. 1.30– 3.30 ,,	Preliminary ·0325 ·0332	Preliminary ·0364 ·0362		— ·0039 ·0030	
Light 3.30 P.M.		3.30 P.M. 5.30 ,,	38·3 38·5	3.30– 5.30 P.M. 5.30– 7.30 ,, 7.30– 9.90 ,, 9.30–11.30 ,,	Preliminary ·0332 ·0337 ·0343	Preliminary ·0156 ·0172 ·0194	— ·0176 ·0155 ·0149	— — — —	— — — —
Darkened 11.30 P.M.	2nd weight, 1·45 grammes.	9.30 A.M.	· 38·3	11.30– 1.30 A.M. 1.30– 3.30 ,, 3.30– 5.30 ,,	Preliminary ·0352 ·0349	Preliminary ·0399 ·0404		— ·0047 ·0055	
				Mean values per hour			·0081½	·0023	·0130

Experiment XLIV.—February 9, 1903.   40°·9 C.

Illumina-tion.	Weight and area.	Temperature.		Time.	CO₂ supplied.	CO₂ in current after leaving chamber.	Apparent assimila-tion per 2 hours.	Respira-tion per 2 hours.	Real assimila-tion per hour per 50 sq. cms.
Darkened.	Area, 41·4 sq. centims. 1st weight, 1·55 grammes.	40°·9 C. throughout experiment.		10.0 –11.30 A.M. 11.30– 1.30 P.M. 1.30– 3.30 ,,	Preliminary ·0335 —	Preliminary ·0360 —		— ·0025 —	
Light 3.30 P.M.				3.30– 5.30 P.M. 5.30– 7.30 ,, 7.30– 9.30 ,, 9.30–11.30 ,,	Preliminary ·0360 ·0370 ·0368	Preliminary ·0237 ·0262 ·0288	— ·0123 ·0108 ·0080	— — — —	— — — —
Darkened 11.30 P.M.	2nd weight, 1·55 grammes.			11.30– 1.30 A.M. 1.30– 3.30 ,, 3.30– 5.30 ,,	Preliminary ·0364 ·0362	Preliminary ·0398 ·0387		— ·0034 ·0025	
				Mean values per hour			·0051½	·0014	·0078

TABLE VII.—*continued.*

Illumination.	Weight and area.	Temperature.	Time.	CO₂ supplied.	CO₂ in current after leaving chamber.	Apparent assimilation per 2 hours.	Respiration for 2 hours.	Real assimilation per hour per 50 sq. cms
			Experiment XLV.—February 10, 1903. 40°·9 C.					
Darkened.	Area, 40·0 sq. centims. Weight, 1·60 grammes.	40°·9 C. throughout experiment.	10.0 –11.30 A.M. 11.30– 1.30 P.M. 1.30– 3.30 ,,	Preliminary ·0354 ·0361	Preliminary ·0379 ·0377		— ·0025 ·0016	
Light 3.30 P.M.			3.30– 5.30 P.M. 5.30– 7.30 ,, 7.30– 9.30 ,, 9.30–11.30 ,,	Preliminary ·0342 ·0349 ·0345	Preliminary ·0241 ·0240 ·0247	— ·0101 ·0109 ·0098	— — — —	— — — —
Darkened 11.30 P.M.			11.30– 1.30 A.M. 1.30– 3.30 ,, 3.50– 5.30 ,,	Preliminary ·0338 ·0333	Preliminary ·0379 ·0379		— ·0041 ·0046	
				Mean values per hour		·0051	·0016	·0080
			Experiment XLVI.—February 6, 1903. 42°·9 C.					
Darkened.	Area, 29·0 sq. centims. 1st weight, 1·48 grammes.	42°·9 C. throughout experiment.	10.30–12.0 noon 12.0 – 2.0 P.M. 2.0 – 4.0 ,,	Preliminary ·0291 ·0294	Preliminary ·0327 ·0319		— ·0036 ·0025	
Light 4 P.M.			4.0 – 6.0 P.M. 6.0 – 8.0 ,, 8.0 –10.0 ,, 10.0 –12.0 night	Preliminary ·0370 ·0387 ·0399	Preliminary ·0328 ·0355 ·0377	— ·0042 ·0032 ·0022	— — — —	— — — —
Darkened midnight.	2nd weight, 1·48 grammes.		12.0 – 2.0 A.M. 2.0 – 4.0 ,, 4.0 – 6.0 ,,	Preliminary ·0398 ·0382	Preliminary ·0423 ·0418		— ·0025 ·0036	
				Mean values per hour		·0016	·0015	·0040
			Experiment XLVII.—February 7, 1903. 42°·9 C.					
Darkened.	Area, 39·2 sq. centims. 1st weight, 1·48 grammes.	42°·9 C throughout experiment.	10.0 –11.30 A.M. 11.30– 1.30 P.M. 1.30– 3.30 ,,	Preliminary ·0395 ·0391	Preliminary ·0425 ·0406		— ·0030 ·0015	
Light 3.30 P.M.			3.30– 5.30 P.M. 5.30– 7.30 ,, 7.30– 9.30 ,, 9.30–11.30 ,,	Preliminary ·0398 ·0412 ·0402	Preliminary ·0372 ·0375 ·0382	— ·0026 ·0037 ·0020	— — — —	— — — —
Darkened 11.30 P.M.	2nd weight, 1·48 grammes.		11.30– 1.30 A.M. 1.30– 3.30 ,, 3.30– 5.30 ,,	Preliminary ·0396 ·0387	Preliminary ·0432 ·0416		— ·0036 ·0029	
				Mean values per hour		·0014	·0014	·0035½

TABLE VIII.

Preliminary assimilation experiments with high intensity of light (L.In. = < 45)·
Keith incandescent compound burner at high gas pressure (8 ins. water).

Weight and area.	Time.	CO₂ supplied.	CO₂ in current after leaving chamber.	Apparent assimilation per hour.	Respiration per hour.	Real assimilation calculated for 1 hour and 50 sq. cms.
Experiment XLVIII.—March 3, 1903. Bath temperature 11° C.						
Weight, 1·50 grammes. Area 36·0 sq. centims.	10.15–11.45 A.M. 11.45–12.45 P.M. 12.45– 1.45 ,, 1.45– 2.45 ,, 2.45– 3.45 ,,	Preliminary ·0380 ·0377 ·0382 ·0382	Preliminary ·0315 ·0317 ·0313 ·0320	— ·0065 ·0060 ·0069 ·0062	— — — — —	— — — — —
		Mean values per hour. .		·0064	·0002*	·0090
Experiment XLIX.—March 2, 1903. Bath temperature 29° C.						
Weight, 1·48 grammes. Area 37·0 sq. centims.	10.15–12.15 P.M. 12.15– 1.15 ,, 1.15– 2.15 ,, 2.15– 3.15 ,, 3.15– 4.15 ,,	Preliminary ·0361 ·0375 ·0384 ·0389	Preliminary ·0195 ·0232 ·0225 ·0214	— ·0166 ·0143 ·0159 ·0175	— — — — —	— — — — —
Darkened	4.15 - 6.15 P.M. 6.15– 8.15 ,, 8.15–10.15 ,, 10.15–12.15 A.M.		Preliminary ·0025 ·0022 ·0024	— — — —	— ·0012½ ·0011 ·0012	— — — —
		Mean values per hour. .		·0161	·0012	·0233
Experiment L.—February 27, 1903. Bath temperature 32° C.						
Weight, 1·35 grammes. Area 37·0 sq. centims.	12.30– 1.30 P.M. 1.30– 2.30 ,, 2.30– 3.30 ,, 3.30– 4.30 ,,	Preliminary ·0324 ·0311 ·0305	Preliminary ·0136 ·0150 ·0155	— ·0188 ·0161 ·0150	— — — —	— — — —
		Mean values per hour. .		·0166	·0010*	·0249
Experiment LI.—February 28, 1903. Bath temperature 38° C.						
Weight, 1·55 grammes. Area 39·3 sq. centims.	11.20–12.45 P.M. 12.45– 1.45 ,, 1.45– 2.45 ,, 2.45– 3.45 ,,	Preliminary ·0380 ·0390 ·0378	Preliminary ·0299 ·0315 ·0308	— ·0081 ·0075 ·0070	— — — —	— — — —
		Mean values per hour. .		·0075	·0019*	·0125

* Calculated from Table I.

TABLE IX.

Further preliminary assimilation experiments with high intensity of light (L.In. = 74) Keith incandescent compound burner at high gas pressure (8 ins. water).

Illumination.	Weight and area.	Time.	CO_2 supplied.	CO_2 in current after leaving chamber.	Apparent assimilation per hour.	Respiration per hour.	Real assimilation per hour per 50 sq. cms.
		Experiment LII.—March 9, 1903. Bath temperature 32° C.					
L.In. = 74.	Area, 36·0 sq. centims. 1st weight, 1·40 grammes. 2nd weight, 1·50 grammes. [Leaf brown at centre.]	11.15–12.45 P.M. 12.45– 1.45 ,, 1.45– 2.45 ,,	Preliminary ·0447 ·0457	Preliminary ·0367 ·0402	— ·0080 ·0055	— — —	— — —
				Mean values per hour	·0068	·0010*	·0108
		Experiment LIII.—March 10, 1903. Bath temperature 32°.					
L.In. = 31.	Weight, 1·10 grammes. Area, 34·8 sq. centims.	11.0 –12.30 P.M. 12.30– 1.30 ,, 1.30– 2.30 ., 2.30– 3.30 ,,	Preliminary ·0420 Spoilt ·0428	Preliminary ·0292 — ·0320	— ·0128 — ·0108	— — — —	— — — —
L.In. = 74.		3.30– 4.30 P.M. 4.30– 5.30 ,, 5.30– 6.30 ,,	·0428 ·0443 ·0439	·0330 ·0367 ·0417	·0098 ·0076 ·0022	— — —	— — —
Darkened. CO_2 free current.	[Leaf one quarter brown.]	6.30– 8.30 P.M. 8.30–10.30 ,, 10.30–12.30 A.M.		Preliminary ·0020 ·0021		— ·0010 ·0010½	
				Mean values per hour	·0086	·0010	·0129
		Experiment LIV.—March 11, 1903. Bath temperature 11° C.					
L.In. = 74.	Weight, 1·55 grammes. Area, 38·7 sq. centims. [Leaf quite green.]	11.0 –12.30 P.M. 12.30– 1.30 ,, 1.30– 2.30 ,, 2.30– 3.30 ,, 3.30– 4.30 ,,	Preliminary ·0483 ·0495 ·0485 ·0490	Preliminary ·0333 ·0358 ·0360 ·0368	— ·0150 ·0137 ·0125 ·0122	— — — — —	— — — — —
				Mean values per hour	·0134	·0002*	·0176

* Calculated from Table I.

TABLE X.

An experiment with a Keith high pressure burner showing the unavoidable rise in the assimilation maximum that results, with each augmentation of the light intensity, from the decided increase of internal leaf temperature that the radiation produces, in spite of the bath temperature being kept constant throughout.

Illumination.	Weight and area.	Probable temperature.*	Time.	CO_2 supplied.	CO_2 in current after leaving chamber.	Apparent assimilation per hour.	Real assimilation per hour per 50 sq. cms.†
			Experiment LV. March 12-15, 1903.				
L.In. = 18·5	Weight, 1·40 grammes. Area 40·5 sq. centims.	18°·5 C.	11.0 -12.30 P.M.	Preliminary	Preliminary	—	·0100
			12.30- 1.30 ,,	·0483	·0398	·0085	
			1.30- 2.30 ,,	·0497	·0426	·0071	
			2.30- 3.30 ,,	·0499	·0422	·0077	
L.In. = 9·25		15° C.	3.30- 4.30 P.M.	Preliminary	Preliminary	—	·0068½
			4.30- 5.30 ,,	·0280	·0220	·0060	
			5.30- 6.30 ,,	·0272	·0225	·0047	
			6.30- 7.30 ,,	Spoilt	Spoilt	—	
L.In. = 4·75		13° C.	7.30- 8.55 P.M.	Preliminary	Preliminary	—	·0055½
			9. 0-10. 0 ,,	·0498	·0453	·0045	
			10. 0-11. 0 ,,	·0505	·0463	·0042	
			11. 0-12. 0night	·0492	·0450	·0042	
			Darkened all night. Relighted 9.40 A.M., March 13.				
L.In. = 4·75		13° C.	9.40-11.10 A.M.	Preliminary	Preliminary	—	·0059
			11.10-12.10 P.M.	·0400	·0350	·0050	
			12.10- 1.10 ,,	·0420	·0375	·0045	
			1.10- 2.10 ,,	·0432	·0390	·0042	
L.In. = 2·87		11° C.	2.10- 3.10 P.M.	Preliminary	Preliminary	—	·0039
			3.10- 4.10 ,,	·0441	·0413	·0028	
			4.10- 5.10 ,,	·0468	·0433	·0035	
			5.10- 6.10 ,,	·0445	·0420	·0025	
L.In. = 1·47		11° C.	6.10- 7.10 P.M.	Preliminary	Preliminary	—	·0024
			7.10- 8.10 ,,	·0447	·0426	·0021	
			8.10- 9.10 ,,	·0447	·0427	·0020	
			9.10-10.10 ,,	·0448	·0432	·0016	
			Darkened for 36 hours. Relighted 9.30 A.M., March 15.				
L.In. = 1·47		11° C.	9.30-11.30 A.M.	Preliminary	Preliminary	—	·0030½
			11.30- 1.30 P.M.	·0335	·0293	·0042	
			1.30- 3.30 ,,	·0339	·0284	·0055	
L.In. = ·73		11° C.	3.30- 4.30 P.M.	Preliminary	Preliminary	—	·0018½
			4.30- 5.30 ,,	·0328	·0307	·0021	
			5.30- 6.30 ,,	·0335	·0305	·0030	
			6.30- 7.30 ,,	·0321	·0293	·0028	

* Based on determinations made later, see p. 76.

† Respiration for respective temperatures allowed from Table I.

TABLE XI.

Experiments showing maximal assimilation at temperatures between 15° C. and 40° C. Keith incandescent compound burner at high gas pressure (8 ins. water), and electrical determinations of internal leaf temperature.

Illumina-tion.	Weight and area.	Temperature.		Time.	CO$_2$ supplied.	CO$_2$ in current after leaving chamber.	Apparent assimila-tion per hour.	Respira-tion per hour.	Real assimila-tion cal-culated for 50 sq. cms. and 1 hour.
		Time.	Temp.						
				Experiment LVI.—April 6, 1903. 15°·0 C.					
L.In. = 13.	Area, 37·0 sq. centims. Weight, 1·50 grammes.	Internal tempera-ture of leaf, 15° C. Temperature of bath, 10° C.		10.30–12.10 P.M. 12.10– 1.10 „ 1.10– 2.10 „ 2.10– 3.10 „ 3.10– 4.10 „	Preliminary ·0356 ·0352 ·0353 ·0360	Preliminary ·0306 ·0306 ·0303 ·0312½	— ·0050 ·0046 ·0050 ·0047½	— — — — —	— — — — —
Darkened 4.30 P.M. CO$_2$ free current.		4.15 P.M. 11 P.M.	15° 0 15°·0	4.10– 6.0 „ 6.0 –11.12 „		Preliminary ·0017		— ·0003½	
					Mean values per hour		·0048½	·0003½	·0070½
				Experiment LVII.—April 4, 1903. 23°·7 C.					
L.In. = 26.	Area, 42·0 sq. centims. Weight, 1·48 grammes.	Temperature of leaf, 23°·7 C. Temperature of bath, 11° C.		12.30– 2.0 P.M. 2.0 – 3.0 „ 3.0 – 4.0 „ 4.0 – 5.0 „ 5.0 – 6.0 „	Preliminary ·0382 ·0418 ·0374 ·0325	Preliminary ·0317 ·0338 ·0290 ·0249	— ·0065 ·0080 ·0084 ·0076	— — — — —	— — — — —
Darkened 6 P.M. CO$_2$ free current.		6.35 P.M. 11.40 „	23°·7 23°·7	6.0 – 7.30 P.M. 7.30– 9.30 „ 9.30–11.30 „		Preliminary ·0020½ ·0015½		— ·0010 ·0008	
					Mean values per hour		·0076	·0009	·0101
				Experiment LVIII.—April 3, 1903. 30°·5 C.					
L.In. = 45.	Area, 46·0 sq. centims. Weight, 1·60 grammes.	Temperature of leaf, 30°·5 C. Temperature of bath, 13°·5 C.		10.0 –11.30 A.M. 11.30–12.30 P.M. 12.30– 1.30 „ 1.30– 2.30 „ 2.30– 3.30 „	Preliminary ·0360 ·0386 ·0386 ·0379	Preliminary ·0226 ·0270 ·0280 ·0280	— ·0134 ·0116 ·0106 ·0099	— — — — —	— ·0157 — — —
Darkened 4 P.M. CO$_2$ free current.		4.50 P.M. 6.0 „ midnight	30°·7 31°·3 31°·5	4.0 – 5.30 P.M. 5.30– 7.30 „ 7.30– 9.30 „ 9.30–11.30 „		Preliminary ·0023 ·0022 ·0024		— ·0011½ ·0011 ·0012	
					Mean values per hour		·0113½	·0011½	·0136

TABLE XI.—*continued*.

Illumination.	Weight and area.	Temperature.		Time.	CO₂ supplied.	CO₂ in current after leaving chamber.	Apparent assimilation per hour.	Respiration per hour.	Real assimilation calculated for 50 sq. cms. and 1 hour.
		Time.	Temp.						

$$\text{Experiment LIX.—April 7, 1903. } 37°\cdot5 \text{ C.}$$

Illumination.	Weight and area.	Time.	Temp.	Time.	CO₂ supplied.	CO₂ in current after leaving chamber.	Apparent assimilation per hour.	Respiration per hour.	Real assimilation calculated for 50 sq. cms. and 1 hour.
L.In. = 45.	Weight, 1·43 grammes. Area, 36·0 sq. centims.	Internal temperature of leaf, 37°·5 C. Temperature of bath, 28°·5 C.		10.30–12.0 noon 12.0 – 1.0 P.M. 1.0 – 2.0 ,, 2.0 – 3.0 ,, 3.0 – 4.0 ,,	Preliminary ·0364 ·0366 ·0384½ ·0409	Preliminary ·0210 ·0260 ·0305 ·0350	— ·0154 ·0106 ·0079½ ·0059	— — — —	— ·0237 — — —
Darkened 4.10 P.M. CO₂ free current.		4.30 P.M. 5.30 ,, 11.30 ,,	37·6 37·4 37·6	4.10– 5.30 P.M. 5.30– 7.30 ,, 7.30– 9.30 ,, 9.30–11.30 ,,		Preliminary ·0040 ·0042 ·0036½		— ·0020 ·0021 ·0018	
					Mean values per hour		·0100	·0019	·0163

$$\text{Experiment LX.—April 9, 1903. } 40°\cdot5 \text{ C.}$$

Illumination.	Weight and area.	Time.	Temp.	Time.	CO₂ supplied.	CO₂ in current after leaving chamber.	Apparent assimilation per hour.	Respiration per hour.	Real assimilation calculated for 50 sq. cms. and 1 hour.
L.In. = 45.	Weight, 1·45 grammes. Area, 38·5 sq. centims.	Temperature of leaf, 40°·5 C. Temperature of bath, 32°·5 C.		2.50– 4.20 P.M. 4.20– 5.20 ,, 5.20– 6.20 ,, 6.20– 7.20 ,, 7.20– 8.20 ,,	Preliminary ·0380 ·0390 ·0390 ·0393	Preliminary ·0281 ·0315 ·0330 ·0373	— ·0099 ·0075 ·0060 ·0020	— — — —	— ·0149 — —
Darkened 8.20 P.M. CO₂ free current.		8.30 P.M. 11.30 ,, 9.30 A.M.	40·6 40·5 40·4	8.20– 9.50 P.M. 9.50–11.50 ,, 11.50– 1.50 A.M. 1.50– 3.50 ,,		Preliminary ·0029 ·0032 ·0035		— ·0014½ ·0015½ ·0017½	
					Mean values per hour		·0063	·0016	·0102